ECL 18
ME 116

OXFORD LABORATORIES

Prothrometer Design

In 1960, Oxford Laboratories was a small new company in the medical instrumentation field. Oxford had been financed by a group of medical doctors. The company had four employees: a general manager, a salesman, a secretary, and an assembly worker. In addition, Oxford planned to use manufacturer's representatives to increase its sales capability. Oxford's facilities included an assembly room, research laboratory, and sales office.

Mr. Leonard Sullivan, General Manager, was confronted by design problems with the Laboratories' first product, called the Oxford Prothrometer. It was an instrument designed for use in anticoagulant therapy by medical doctors and technicians. It was used to determine "prothrombin time", the period required for coagulation to occur after a patient's whole blood was mixed with a specially prepared reagent. The prothrombin time indicated how much anticoagulant medication was in the patient's blood. This data was used by the doctor for prescribing additional treatment. The instrument and its operation are described in greater detail in Exhibit (1).

The Prothrometer had been designed by a "moonlighting" draftsman. Mr. Sullivan commented that although he was confident of the product concept, he had lost faith in the product itself. Shortly after initiating its sales in February, 1960, Oxford received complaints from customers about the operation of the instrument. Mr. Sullivan commented that many Prothrometers had been returned to Oxford because of misalignment of the timer-motor assembly. The returned instruments resulted in loss of reputation for Oxford and inconvenience for its customers.

Mr. Sullivan also noted that with the present design, (1) the instrument was difficult, and therefore, expensive to assemble, (2) the sheet metal case was expensive to manufacture, (3) the instrument did not look its price. The timer-motor assembly, sheet metal case, and other components are noted in the assembly drawing included in Exhibit (2).

Prepared in the Design Division of the Mechanical Engineering Department by J. Kendall Williams under the direction of Robert H. McKim.

In mid-1960, Oxford began advertising in San Francisco Peninsula newspapers for medical inventions in the hope of expanding its product line. A Stanford undergraduate student read this ad in the Stanford Daily and informed one of his instructors, Mr. Robert H. McKim, of Oxford's interests. Mr. McKim had developed an invention related to the medical market, and he contacted Oxford Laboratories.

Although Mr. McKim's discussion with Oxford did not lead to the production of his invention since it did not fall within Oxford's marketing capability, Mr. Sullivan was able to direct Mr. McKim's attention to the production problems of the Prothrometer. During one of their meetings, Mr. Sullivan demonstrated the use of the Prothrometer and explained the nature of the production problems. He noted at that time that the current inventory of instruments would be exhausted in three months. This meant that revised production drawings would have to be completed in five weeks if Oxford was to meet its delivery commitments with an improved model.

Exhibit 1. Oxford Prothrometer.

Exhibit 1. Oxford Prothrometer, Con't.

Exhibit 1. Oxford Prothrometer, Con't.

Fast, accurate, micro prothrombin times

with the Oxford Prothrometer®

Automatic temperature regulator

Self contained chronometer

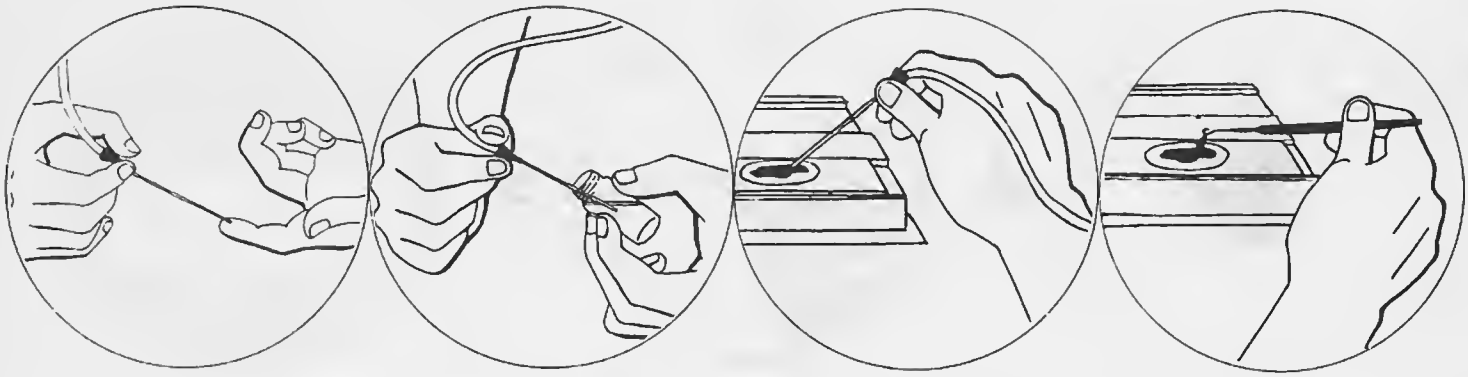
assures reliable anticoagulant therapy control with micro techniques using: capillary whole blood, or capillary plasma without centrifugation,* or capillary whole blood clotting time*†... at low cost.

*Weiner, M. and Messinger, W., Microclotting Techniques, *J. Lab. Clin. Med.* In press.

†Lewis, R. C. and Glueck, H. I., A Micromethod for Determining Clotting Time, Using Capillary Blood and Siliconized Tubes, *J. Lab. Clin. Med. 52:* 299, 1958.

Immediate, reproducible determinations in

four simple steps:



1. Take measured volume of blood from finger puncture or earlobe

2. Draw same measured volume of thromboplastin or reagent

3. Expel blood with reagent into slide dimple and start timer

4. Mix and stop timer when coagulum or clot appears

Ordering information:

Cat. #123 Oxford Prothrometer kit, complete, including: M 103 Prothrometer (115v, 60 cycle), thermometer, pipettes, stirring device, slides, lancettes, spot bandages and carry case, $148.50.

Cat. #124 Same as #123 without carrying case, $138.50.

Cat. #125 Oxford M 103 Prothrometer only, without accessories, $118.50.

Cat. #511 75 mm premarked disposable capillary pipettes, per 100, $2.75.

Prices for other accessories available on request.
All prices F.O.B. Redwood City, California and subject to change without notice.

For further information, see our authorized distributor, or write

Oxford Laboratories®

961 Woodside Road, Redwood City, California

FORM 2M—360

OPERATING INSTRUCTIONS FOR THE OXFORD PROTHROMETER® ME-116

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EMerson 8-9138

The use of oral anticoagulants for prevention or contral of thrambaembolic or thrombotic incidents has now been reported in clinical studies involving thousands of patients.[1] Such studies indicate that oral anticoagulants reduce mortality as much as fifty percent in the first six weeks after myocordial infarction. Similarly, long-term aral onticoagulant therapy has diminished extension of the thrombotic process and has decreased mortality.[2]

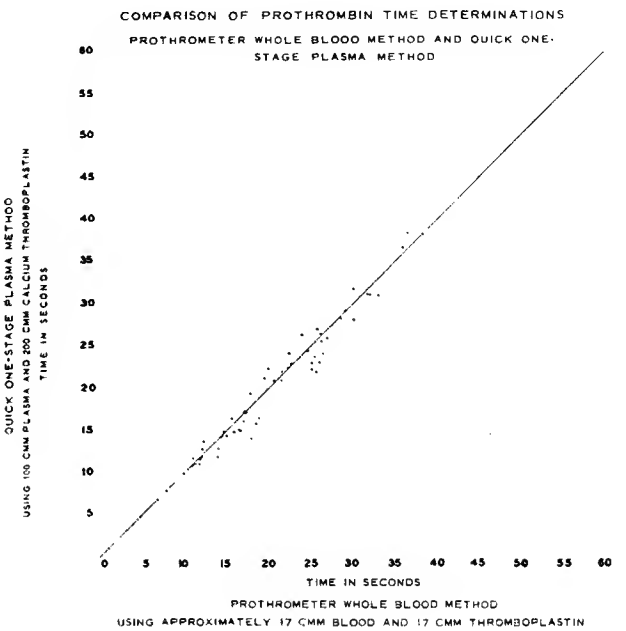
Since the discovery of heparin (see reference 3) and bishydraxycoumarin (Dicumarol®) by Link[4] in 1939, many tests have been developed to determine prothrombin activity. Such tests are necessary for adequate and safe administratian af anticaagulants. The tests must yield results which, when praperly interpreted, pravide the clinician with the basis far regulating anticoagulant dosage while abtaining maximum protectian against thrombasis with minimum risk of hemorrhage.

The Prothrameter has been designed to permit fast and accurate (see scattergram at right) estimation of "prothrombin time", using minute amaunts of finger-puncture whole blood. Control of anticoagulant therapy is simplified when the Prothrometer is employed because the physician can obtain "prothrombin time" values in a very few minutes. In addition, cost of each determination is considerably reduced since the amount of thramboplastin reagent required is appreciably less than called for in traditional macro methods.

The following techniques may be performed with the Prothrometer:

1. Whole blood "prothrombin time" employing finger puncture blood and aerformed with premarked disposable capillary "pipettes".
2. Whole blood "prothrombin time" employing finger puncture blood and performed with 20 cmm Sohli-type glass pipettes.

These methods have been carefully tested and have each proven satisfactory for controlling anticoagulent therapy in large numbers of treated patients.



BIBLIOGRAPHY:

1. Gilchrist, A. R. and Tulloch, J. A., "Anticoagulants in caronary diseases", Brit. Med. J., 2:720-724, 1954.

2. Friedburg, C. K., "A critical appraisal of anticoagulants for short-term and long-term use in the manogement of myocardial infarction and systemic arterial embolism--Part II", N. Y. St. J. Med., 58:1303-1308, 1958.

3. Howell, W. H. and Holt, E., "Two new factors in blood coagulation - heparin and pro-antithrombin", Am. J. Physiol., 47:328-341, 1918-1919.

4. Link, K. P., "The onticoagulant dicumarol", Proc. Inst. Med. Chicago, 15:370-389, 1945.

5. Jacques, L. B., "Estimation of the blood prothrombin by the bedside method", in "The coagulatian af blood--methads af study", edited by Tocantins, L. M., Grune and Stratton, N. Y., 1955.

6. Goldfeder, A., Bloom, D., and Weiner, M., "A new improved method for determination of prothrombin levels in blaod", Science, 111:365, 1950.

7. Manchester, B., "The value of continuous (1 ta 10 years) long-term anticoagulant therapy", Ann. Int. Med., 47:1202-1209, 1957.

8. Haffman, O. D. and Custer, R. P., "A micro method for determining prothrombin time on fresh copillary blaad using standard physical conditions", Amer. J. Med. Sci., 204:420-430, 1942.

9. Weiner, M. and Messinger, W. J., "Microclotting techniques", J. Lab. and Clin. Med., October 1960, in press.

10. Miale, J. B. and Winningham, A. R., "A true micromethod for prothrombin time, using capillary blood and disposable multipurpose micropipete", Am. J. Clin. Path., 33:214-219, 1960.

11. Reich, C., "The feasibility of using finger blaod far prothrombin determinations", Am. J. Clin. Path., 30:19, 1958.

SUGGESTIONS AND PRECAUTIONS:

Eject blood and reagent as near the center of the slide concavity as possible so that test will be performed at deepest point in dimple.

It is very important that finger punctures be deep so that blood flows freely. Squeezing should be minimal in order to prevent contamination af the sample with tissue thromboplastin.

If the tip of the finger is cool and relatively ischemic, it should be placed in warm water for a few moments and dried before puncture. Ear lobe blaod may be used in lieu of finger puncture blood.

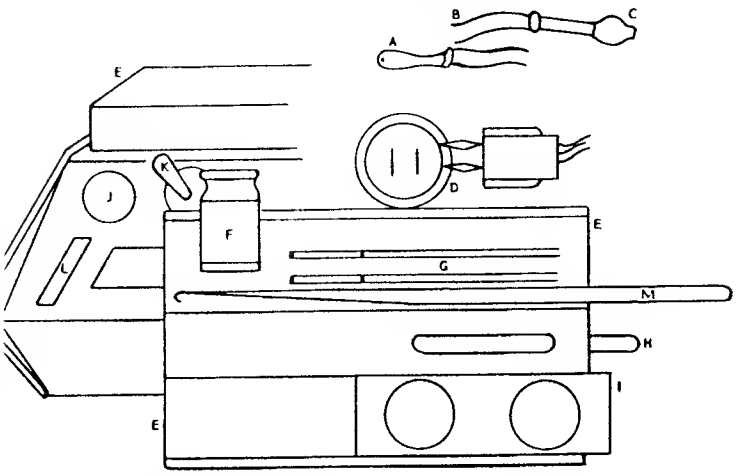
The activity of thromboplastin deteriarates with time. Failure to check with normal controls may allow the use of deteriorated thromboplastin with resulting spuriously long prothrombin times. Follow reagent manufacturer's directions for reconstituting thromboplastin, checking controls, and storage of solutions. Nate: Numerous manufacturers affer lyophylized, pooled human plasma control preparations for evaluating thromboplastin reagents. It is important to remember that such preparations are satisfactory when employed salely for determining whether a particular thromboplastin reagent is within a satisfactory range of activity. Percent normal activity on test plasma may be derived from such determinations. When whole-blood (as with the Prothrometer) instead of plasma methods are used, a test should be performed on whole-blood fram a so-called "narmal" individual if it is desired to report results in terms of percent normal activity as opposed to actual time in seconds.

If the temperature of the heating block of the Prothrameter varies more than 37.5° to 40° C., contoct your deoler for service.

It is recommended that the technique of choice be practiced a sufficient number of times in order that consistent reproducible results are obtained. For this purpose, lyophylized, pooled human plasma contral preparatians are satisfactory in lieu of finger puncture blood.

ECHNIQUE #1 DISPOSABLE "PIPETTE" METHOD.

A. Mauthpiece	G. Premarked dispasable capillary "pipettes"
B. Rubber tubing	H. Thermameter
C. Bulb adapter	I. Spot slide
D. 115 Volt, A.C., 60 cycle autlet	J. Pilat light
E. Temperature cantralled black	K. Timer switch
F. Thromboplastin	L. Timer reset
	M. Clot Hoak

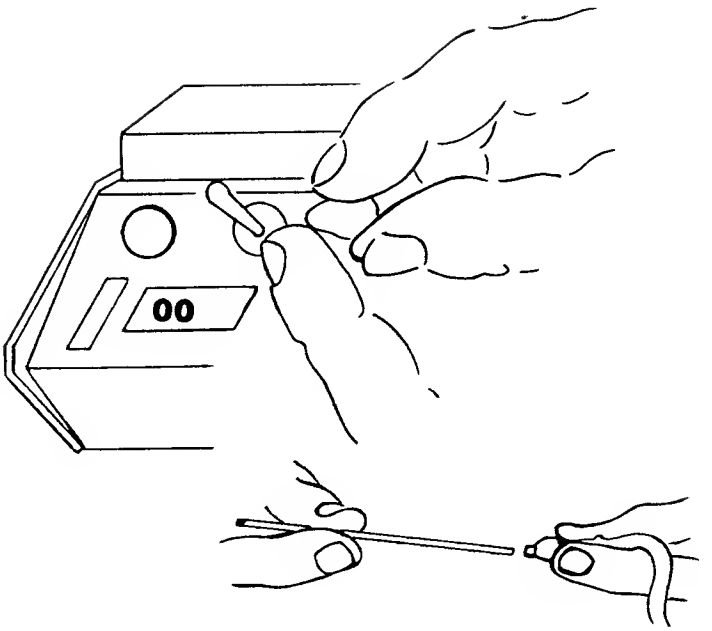


A. Plug Prathrometer into a 115 volt 60 cycle A.C. outlet. (Do not use with D.C.)

B. Place spot slide, clot hook and premarked disposable capillary "pipettes" on top of temperature controlled block, together with small bottle of thromboplastin.

C. Place thermometer in side hole in central portion of block.

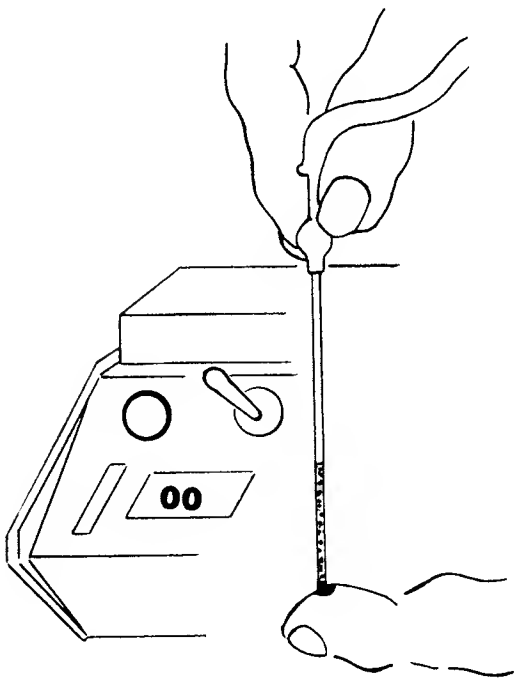
D. Allow reagent o few minutes to heat after block temperature reaches 37.5° to 40° C. (Proper testing range).

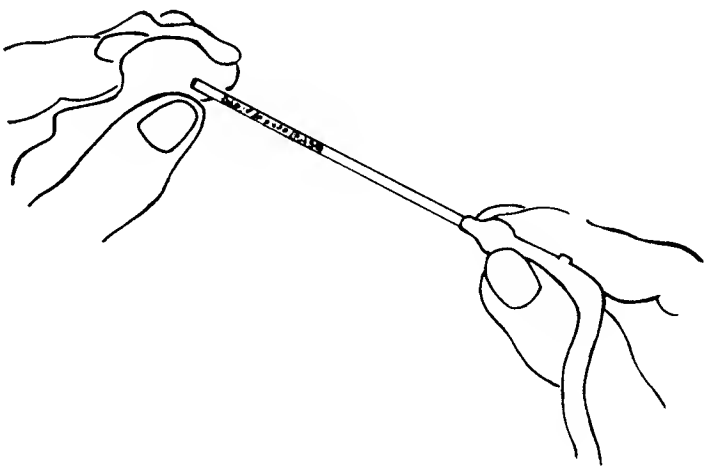


2. A. Clean finger tip or ear lobe with 2" x 2" gauze or cotton saturated in alcohol.

B. Wipe cleaned area dry with sterile 2" x 2" gauze or cotton.

C. Insert warmed premarked disposable capillary "pipette". (Colored tip away from bulb adapter.)



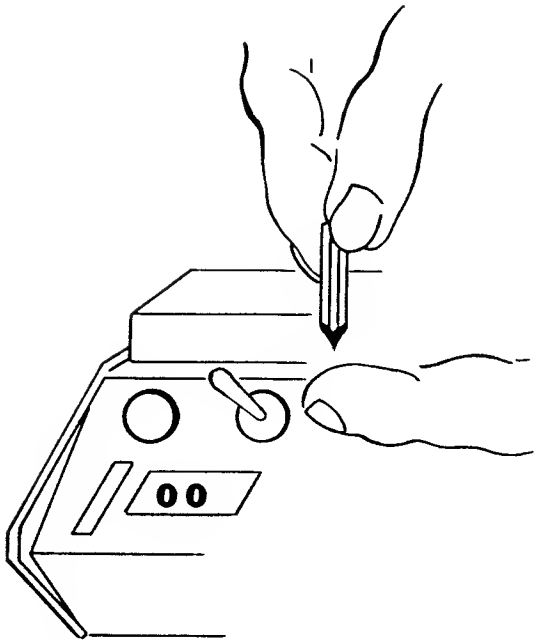


5. A. Slight squeezing to form next drop may be necessary.

B. Using mouthpiece, rubber tubing, and bulb adapter, draw blood inta disposable capillary "pipette" to mork (25 mm from fire polished colored end). Volume = oppraximately 17 cmm.

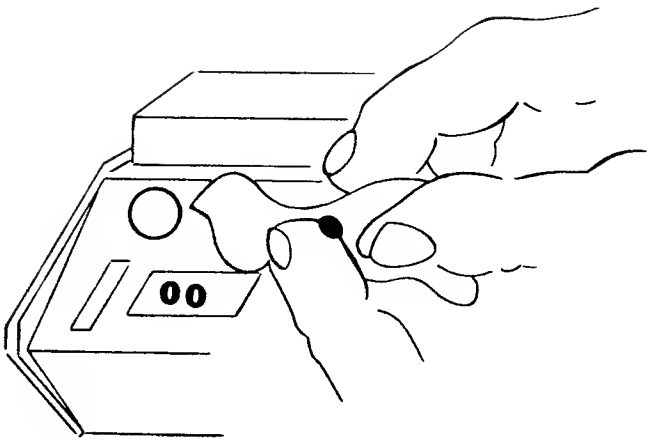
6. A. Tilt "pipette" slightly so thot o small (6 mm) air space is formed at fired end.

B. With tissue, wipe excess blood from outside end of "pipette".

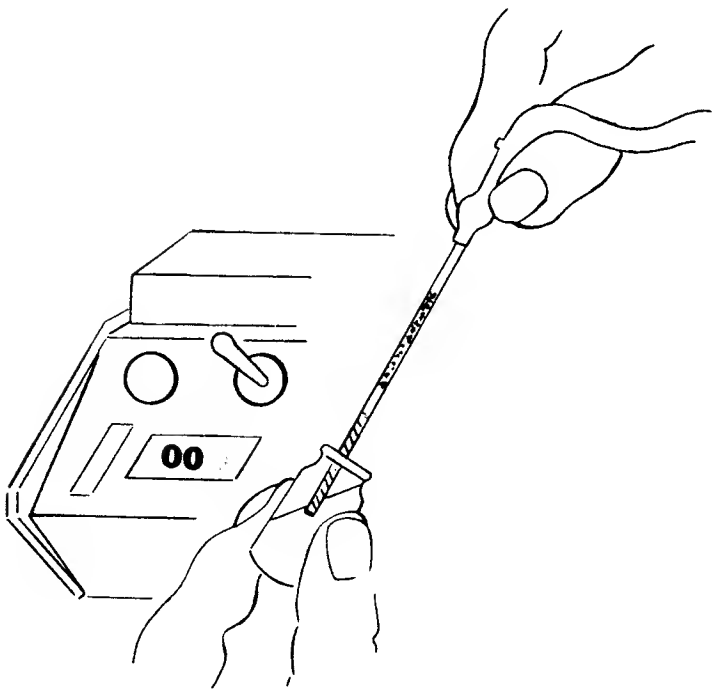


3. Puncture finger tip or ear lobe with lancet.

IMPORTANT: Finger punctures must be sufficiently deep to permit free flow of blood.

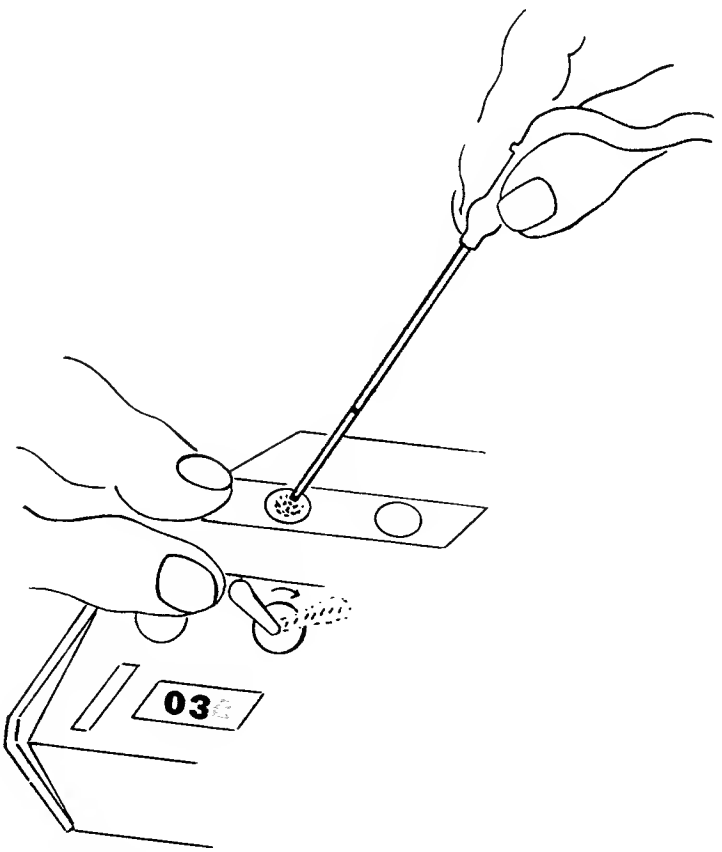


4. Wipe away first drop af blood with sterile 2" x 2" dry gauze or cotton.



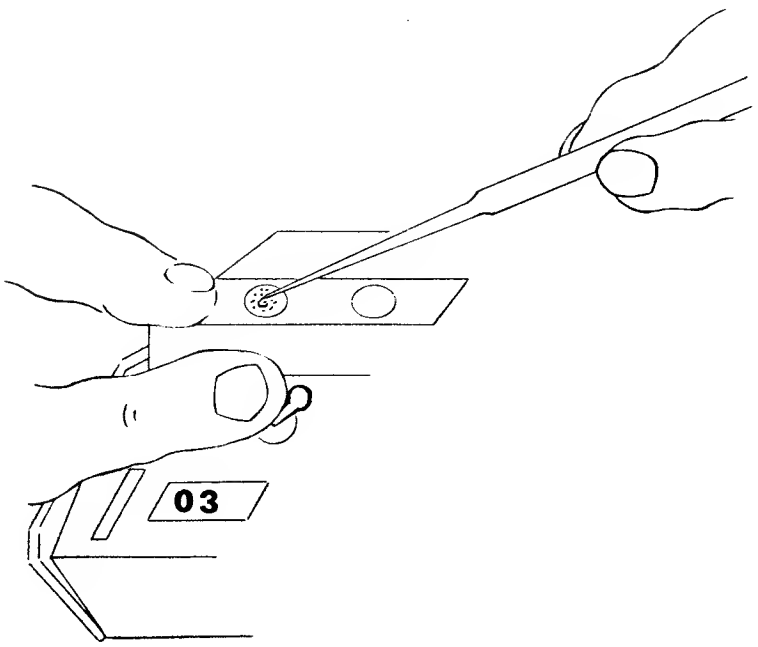
7. Following step 6, next draw warmed thromboplastin behind blood to the same mark on capillary "pipette".
Volume = approximately 17 cmm.

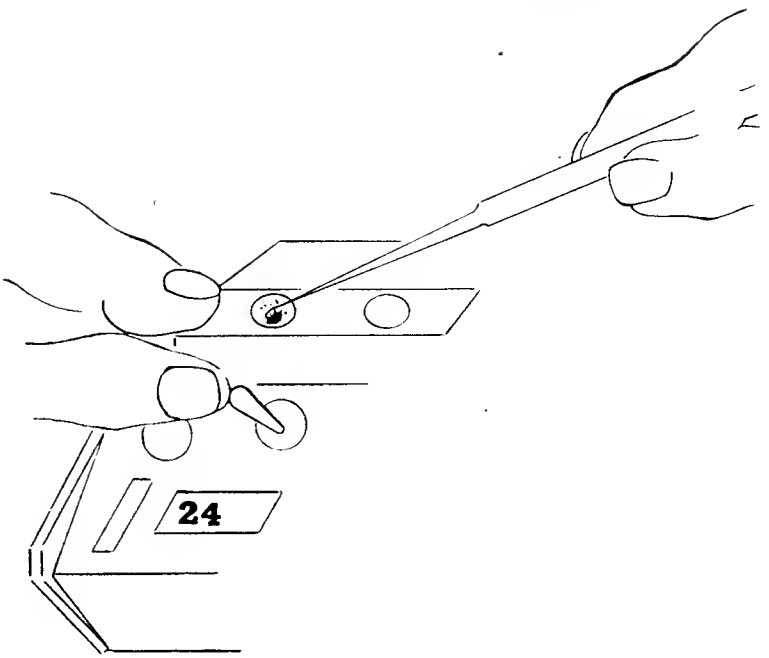
IMPORTANT: Maintain air space between blood and thromboplastin. Reagent must not mix with blood while in premarked disposable capillary "pipette".
If mixing should occur, discard capillary "pipette" and restart pracedure from step #2.



8. Eject entire contents of capillary tube into center af slide concovity and start timer.

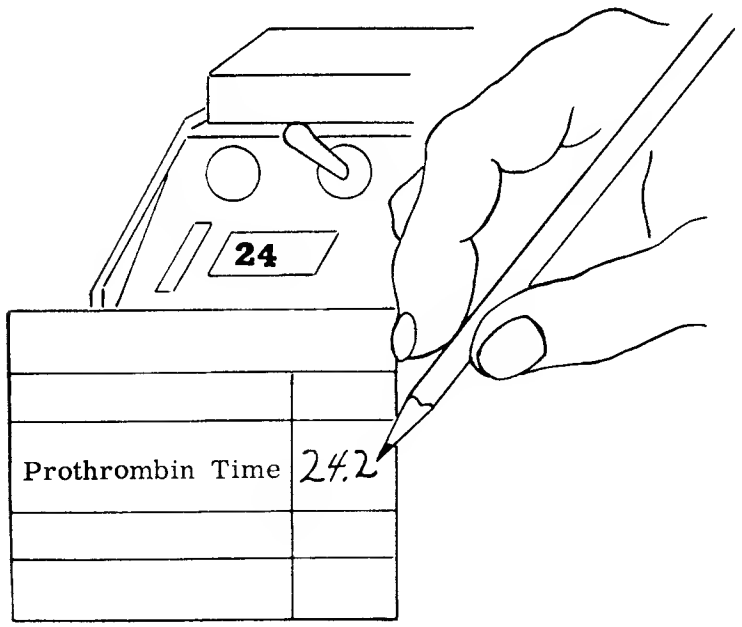
Note: Timer is started the instant all blood and reagent ore ejected inta slide cancavity.





9. A. Immediately pick up special clot hook in right hand.
   B. Stir reagent and blood mixture rapidly in a circular motion.
   C. Leave left thumb on timer switch so that switch can be stopped promptly.

10. Stop timer immediately when coagulum appears.



11. A. Record prothrombin time to nearest 0.1 second.
   B. Slide and clot hook are cleaned with distilled water, alcohol and acetone in that order. Detergents should not be used for cleaning.
   C. A duplicate determination may be run by starting at step #4 providing blood flow is free so that milking is unnecessary. If blood does not flow freely from 1st puncture, it will be necessary to run the duplicate starting at step #2.

TECHNIQUE #2 GLASS PIPETTE METHOD.

1. Plug Prothrometer into a 110 volt 60 cycle A.C. outlet. Place spot slide and clot hook on top of anodized aluminum block together with small bottle of thromboplastin. Place thermometer and pipettes in holes provided in side of heating block. Allow reagent a few minutes to heat after heating block temperature reaches 37.5° to 40° C. (Proper testing range).
2. By means of a warmed pipette, 20 cmm of thromboplastin is placed in one of the concavities of the spot slide.
3. Puncture finger tip or ear lobe with lancet.
4. Using a second warmed pipette, draw 20 cmm of blood and eject into the thrombaplastin in the slide concavity. Simultaneously, start timer.
5. Stir reagent and blood mixture rapidly in a circular motion with special clot hook.
6. Stop timer when coagulum appears.
7. Record prothrombin time to 0.1 second.
8. Slide, clot hook, and pipettes are cleaned with water, alcohol and acetone in that order. Detergents should not be used for cleaning.

Accessories for the Prothrometer, such as slides, clot hook, thermometer, and premarked disposable capillary "pipettes", may be ordered from your surgical supply dealer, or write:

Oxford Laboratories
961 Woodside Road
Redwood City, Calif.

SUPPLEMENT

OPERATING INSTRUCTIONS FOR THE OXFORD PROTHROMETER®

OXFORD LABORATORIES
961 Woodside Road
Redwood City, California

Enclosed with each Prothrometer are illustrated operating instructions, an article preprint showing comparative studies of the macro plasma and micro whole blood methods, and a booklet describing the use of one type oral anticoagulant. Whole blood methods described for use with the Prothrometer have been carefully checked and correlated under rigid laboratory control. The methods compare favorably with the Quick plasma (macro) method provided both tests are performed properly. The micro techniques are easily performed, but their simplicity does not eliminate the necessity for adherence to obvious, but easily overlooked sound laboratory manipulations.

Before performing any tests, familiarize yourself completely with the operation of the Prothrometer. It is important that you learn how to operate the switch so that this manipulation may be performed easily and rapidly. The index finger of the left hand may be used to hold the slide in place and also as a lever for the left thumb as the switch is actuated. These two fingers should remain on the slide and switch respectively, during performance of the prothrombin test.

Ten to fifteen minutes are required to heat the instrument to the proper temperature, and a thermometer is provided so that this temperature may be checked. The Prothrometer is assembled with the finest micro-thermoswitch and is carefully preset at the factory. However, rough handling during shipment occasionally upsets this setting, in which case the instrument must be returned to us for repair.

After you feel you are sufficiently familiar with the operating mechanism of the instrument, you are ready to start practising a few tests as outlined in your enclosed operating instructions.

REAGENTS

1. Oxford Laboratories do not recommend one brand of thromboplastin reagent over another. Simplastin, Calsoplastin, Permaplastin, Acuplastin, and Soluplastin have been tested by us and have been found suitable with the methods described. We are certain other commercially available reagents are equally satisfactory. However, for best results the same brand of reagent should be used from day to day. For information concerning a specific reagent, please write, providing all the information you have about the reagent in question.

2. Regardless of which brand you choose, reconstitution of the thromboplastin must be performed exactly according to the reagent manufacturer's directions. If dehydrated calcium thromboplastin such as Simplastin, Calsoplastin, or Acuplastin is used, the reagent must be reconstituted with distilled water of good quality. The pH should not be lower than 6.0 and the temperature should not exceed 37° C.

3. Transfer pipettes used to measure the distilled water must be clean and dry, and preferably should be used for this one procedure.
NOTE: Two varieties of transfer pipettes are available - "To Deliver" and "To contain." These are usually identified by the markings "TD" or "TC". If no markings are visible, the "To Contain" or "blowout" pipettes are frosted at the top, whereas "To Deliver" pipettes are clear. It is important that you are familiar with the proper method of handling the pipettes you have so that erroneous measurements will not occur.

4. Most reagent manufacturers also distribute lyophilized plasma preparations which are suitable for establishing the normal potency for thromboplastin. Reconstitution of this plasma should be accomplished as is directed by the manufacturer. Do not use the same transfer pipette for reconstituting both plasma and thromboplastin, as contamination of the plasma, thromboplastin, and/or distilled water may result.

5. Establishing the thromboplastin "normal" is accomplished either by using whole blood from "normal" individuals, or by testing the thromboplastin with lyophilized plasma preparations. If the latter is used, the plasma should be preheated on top of the Prothrometer prior to running the test. "Normals" are established in the same manner the regular prothrombin test is run except that plasma preparations as mentioned above may be used in lieu of whole blood. The average of at least three reproduced determinations should be used as the "normal" for the thromboplastin on the day it is reconstituted unless your results appreciably vary from the established "normal" range indicated by the literature enclosed with the reagent. If the plasma preparation is refrigerated immediately after checking the thromboplastin, it should remain stable for 24 hours, so that the same plasma may be used to establish a "normal" on two successive days.

6. Variations from approximately "anticipated normals" of the reagent occur only when

(a) the technique is improperly performed;

(b) the plasma preparation is improperly reconstituted or refrigerated; or

(c) the reagent is improperly reconstituted or has deteriorated.

If such variations occur repeatedly, carefully check each of the aforementioned possibilities until the source of the problem is learned. When reagents and plasma preparations are carefully selected and subsequently handled properly, erroneous determinations rarely result.

7. Most original workers in the anticoagulant field reported results in "per cent of normal" rather than actual seconds, which has needlessly complicated the interpretation of results. Although it is unnecessary to calculate per cent prothrombin activity to regulate dosage properly, (the direct relation between normal and maintenance time in seconds is all that is required), the curves supplied with your reagent are suitable to arrive at "per cent" if desired.

THE TECHNIQUE

1. The Prothrometer kit contains a rubber tubing assembly equipped with a plastic mouthpiece at each end. A small rubber vaccination bulb is stretched tightly over the end of one of the plastic mouthpieces to serve as an adapter to hold the capillary tubes. The capillary tube is inserted through the rubber bulb so that the tube just penetrated the mouthpiece, thereby assuring that the complete assembly is open for suction. If you are inexperienced at using capillary tubes for pipetting, it would be preferable to practice filling the tubes prior to running tests. Actually, very little suction is required, as the capillary tubes usually fill by capillary action. With a little practice, you will be able to regulate quickly the filling of the tubes with blood and reagent.

2. The finger should preferably be prewarmed with warm tap water and wiped dry prior to sticking to assure a free flow of warm blood. If squeezing is necessary, this should be accomplished as near the hand as possible. Excessive squeezing near the puncture site may contaminate the blood sample with tissue thromboplastin.

3. After filling the premarked capillary tube with the proper blood and reagent samples, the contents should be expelled into the center of the slide concavity and the timer started. At this point, it is preferable to release the rubber tubing assembly so that the right hand can be free to stir the mixture. The experienced technician releases the tubing from his hand, but continues to hold onto the assembly in his mouth, which of course, speeds up this procedure considerably.

4. The mixture should be thinned down by stirring outwardly in the slide concavity, thus assuring easier detection of the end point. NOTE: The entire thromboplastin blood mixture does not clot when the end point occurs. The size of the clot formation will vary slightly with different blood samples, but is usually about three times the size of the loop in the clot hook.

CORRELATING RESULTS

The Prothrometer whole blood method is designed to facilitate control of oral anticoagulant therapy, the important comparison therefore being with the previous determination performed with the Prothrometer on the same patient, rather than a comparison with other methods. However, in standardizing your technique, you may initially want to compare your results with results of the method you have been using.

1. It is important to remember that in running a correlation of two methods, both methods must be run at the same time, using the same reagent. It is also preferable that the same sample of blood be used so that the patient is not stuck more than is necessary. Therefore, it is recommended that whole venous blood mixed as follows:

4.5 cc blood to 0.5 cc of 0.1 M sodium oxalate, or

4.5 cc blood to 0.5 cc of 0.1 M sodium citrate

be used when comparing the micro whole blood method with the macro plasma method.

2. The micro method should be run first and immediately after blood is drawn by obtaining the whole blood sample directly from the test tube in which the oxalated or citrated blood is stored. The test tube should be tilted so that the capillary tube may be inserted into the blood without admitting air bubbles into the capillary tube. If for any reason, the micro test is not run immediately after blood is drawn, provision should be made to maintain the blood at 37° C.

3. Although very close correlations are reported in published papers, variations of one or two seconds are probably of no practical significance when comparing one method against another. The important consideration is to determine whether coagulation takes place in the therapeutic range or in the maintenance range.

Oxford Laboratories

Exhibit 2. Oxford Prothrometer Assembly.

ECL-18
M.E. 116

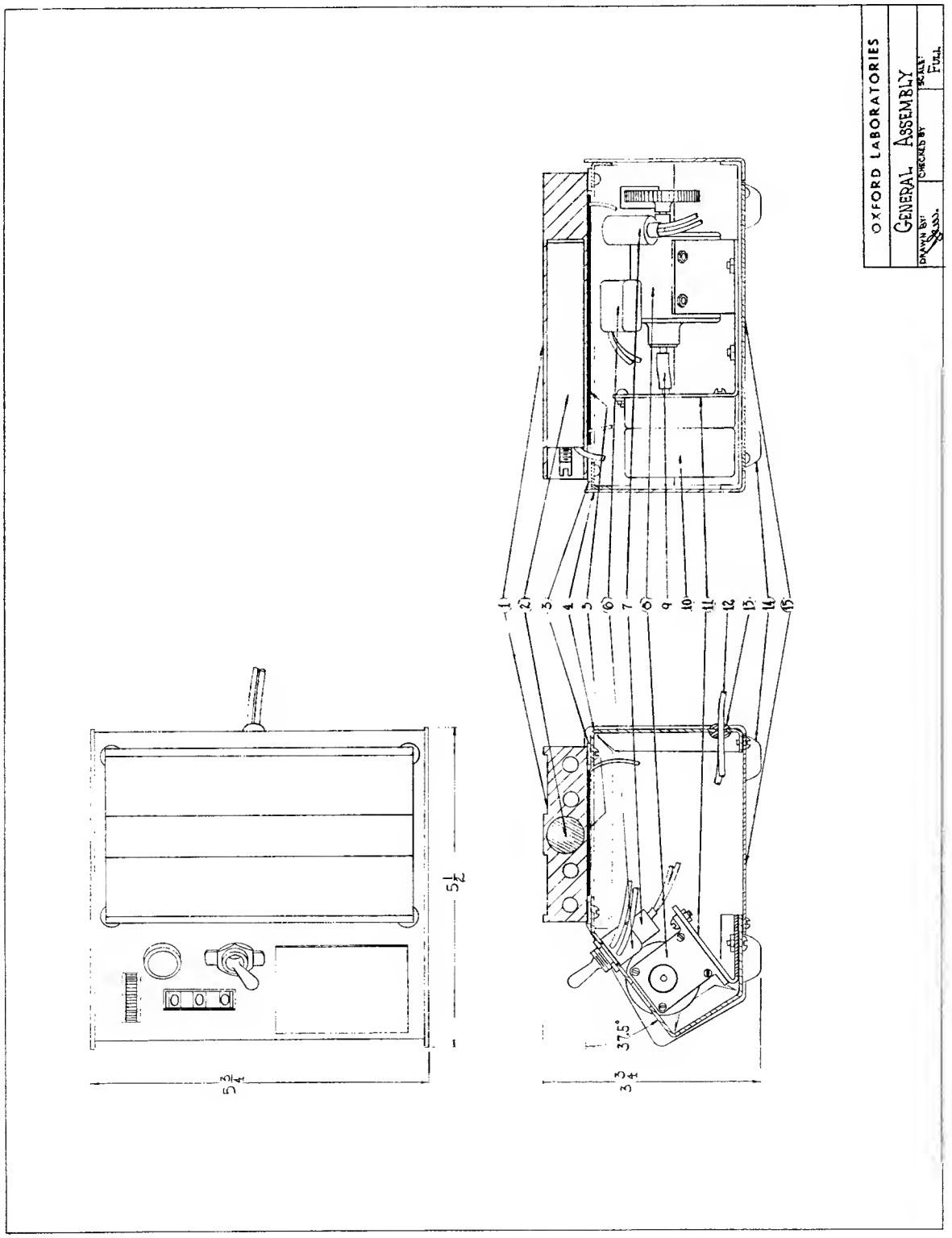


Exhibit 2. Oxford Prothrometer Assembly, Con't.

Assembly Parts List

No.	Description	Req'd
1	Heat Block	1
2	Thermostat	1
3	Washers, Copper	4
4	Case, Sheet Metal	1
5	Heating Pad	1
6	Timer Toggle Switch	1
7	Heat Block Pilot Lamp	1
8	Counter	1
9	Flexible Coupling	1
10	Synchronous Motor	1
11	Motor-Counter Bracket	1
12	Power Cord, 110V, A.C.	1
13	Grommet, Rubber	1
14	Feet, Rubber	3
15	Bottom Plate	1
-	Pan Head Machine Screws 4-40 UNF x 3/16	15
-	Hex Nuts, 4-40 UNF	4
-	Pan Head Machine Screws 10-32 UNF x 1/4	2
-	Hex Nuts, 10-32 UNF	2
-	Decal	1

ECL-18
ME-116

INSTRUCTOR'S MANUAL

Oxford Prothrometer Design

I. Introduction

In most of the situations that a designer faces, the problem is not neatly pre-packaged. Instead, it is usually poorly stated by the client at the outset and gradually expanded and redefined. This was the circumstance confronted by Mr. Robert H. McKim, a design consultant in 1961 for Oxford Laboratories, San Mateo, California. The Oxford Prothrometer case study has been organized to allow the student to assume the role of the design consultant. The purpose of this manual is to provide information that will allow the instructor to assume the role of the client. In addition, material that was not known by either the designer or the client at the outset of the problem is included in the case. This case may be used as desired and is not suggested as the only way to approach the design problem.

The case has been divided into three parts: (1) the "Prothrometer Design", (2) the data "Notes...", and (3) the "Instructor's Manual". The Prothrometer Design packet is organized to provide the students' initial exposure to the Prothrometer design. The material in the Instructor's Manual and data Notes was gradually acquired by Mr. McKim during his design study. It is suggested that the students get this information from the instructor in the same manner - by asking intelligent questions. One of the designer's most important tools is knowing when to ask questions. It is suggested that the data Notes be handed out only when the student requests additional information. Questions that are not answered by one of the data Notes may be resolved by class discussion or outside research.

The data given in the case are included in order to accelerate the learning process, freeing the student from the time-consuming task of gathering his own information. While data collection is a valuable experience, it can be an inefficient process which requires the student to spend time often better used creatively. The student must select the pertinent data from the supplied information. In this respect, this case accelerates data collection by providing more data than the student can use, while not specifying which data are relevant.

Prepared in the Design Division of the Mechanical Engineering Department by J. Kendall Williams under the Direction of Robert H. McKim.

II. Prothrometer Design

In 1960, Oxford Laboratories was a small new company in the medical instrumentation field. Oxford had been financed by a group of medical doctors. The company had four employees: a general manager, a salesman, a secretary, and an assembly worker. In addition, Oxford planned to use manufacturer's representatives to increase its sales capability. Oxford's facilities included an assembly room, research laboratory, and sales office.

Mr. Leonard Sullivan, General Manager, was confronted by design problems with the Laboratories' first product, called the Oxford Prothrometer. It was an instrument designed for use in anticoagulant therapy by medical doctors and technicians. It was used to determine "prothrombin time", the period required for coagulation to occur after a patient's whole blood was mixed with a specially prepared reagent. The prothrombin time indicated how much anticoagulant medication was in the patient's blood. This data was used by the doctor for prescribing additional treatment. The instrument and its operation are described in greater detail in Exhibit (1) of this manual.

The Prothrometer had been designed by a "moonlighting" draftsman. Mr. Sullivan commented that although he was confident of the product concept, he had lost faith in the product itself. Shortly after initiating its sales in February, 1960, Oxford received complaints from customers about the operation of the instrument. Mr. Sullivan commented that many Prothrometers had been returned to Oxford because of misalignment of the timer-motor assembly. The returned instruments resulted in loss of reputation for Oxford and inconvenience for its customers.

Mr. Sullivan also noted that with the present design, (1) the instrument was difficult, and therefore, expensive to assemble, (2) the sheet metal case was expensive to manufacture, (3) the instrument did not look its price. The timer-motor assembly, sheet metal case, and other components are noted in the assembly drawing included in Exhibit (2).

In mid-1960, Oxford began advertising in San Francisco Peninsula newspapers for medical inventions in the hope of expanding its product line. A Stanford undergraduate student read this ad in the Stanford Daily and informed one of his instructors, Mr. Robert H. McKim, of Oxford's interests. Mr. McKim had developed an invention related to the medical market, and he contacted Oxford Laboratories.

Although Mr. McKim's discussions with Oxford did not lead to the production of his invention since it did not fall within Oxford's marketing capability, Mr. Sullivan was able to direct Mr. McKim's attention to the production problems of the Prothrometer. During one of their meetings, Mr. Sullivan demonstrated the use of the Prothrometer and explained the nature of the production problems. He noted at that time that the current inventory of instruments would be exhausted in three months. This meant that revised production drawings would have to be completed in five weeks if Oxford were to meet its delivery commitments with an improved model.

III. Suggested Areas of Interest

A. Cost Estimate for Design Services

Mr. Sullivan had never worked with a design consultant prior to his contact with Mr. McKim. To inform Mr. Sullivan how he intended to approach the Prothrometer design problems, Mr. McKim organized a "design proposal" which noted the time allotted for each phase of work. Under Mr. McKim's contract, he was paid at the end of each phase, and a decision was made at that time whether or not to continue with the next phase.

The student may arrive at a fee for his design services by setting an hourly fee for his time. Mr. McKim noted that $15 per hour was an average figure in the San Francisco Bay area at this time.

Proposal writing is as much a challenge as designing itself. The well-written proposal educates the prospective client about the design services he will receive. Clients who have not employed a designer before are very insecure in this area. Some hope that a few hours of sketching time will produce the necessary "pretty picture". Most want to know detailed information about the proposed work. A poorly organized, ungrammatical, or badly misspelled proposal reflects doubt upon the intelligence and professional capability of the designers. What good is design ability which does not find employment?

The outline which follows indicates a typical student proposal, although it is suggested that the student organize his own proposal form. The data Note that is associated with each part of Phase I of the proposal is indicated in parenthesis below that part. The time allotted for each part may be suggested by the instructor.

SAMPLE STUDENT PROPOSAL

Phase I. RESEARCH AND ANALYSIS	TIME ALLOTTED
A. Background and Application of Blood Test (Student Data: Note on Anticoagulant Therapy)	.....hrs.
B. Human Functions	
1. Operations Analysis (Data: Note on Blood Test Sequence)	.....hrs.
2. Examine Laboratory Environment	.....hrs.

C. Technical Functions

1. Review Nature of Blood Test
(Data: Note on Prothrombin Time Test) .....hrs.

2. Analyze Current Technical Difficulties
(Data: Note on Assembly Problems) .....hrs.

3. Determine Itemized Cost of Present Design
(Data: Note on Manufacturing Costs) .....hrs.

4. Review Materials and Process for New Instruments .....hrs.

5. Conduct Components Search
(Data: Note on Component References) .....hrs.

D. Market Survey

1. Review Merits and Demerits of Competitive Products
(Data: Note on Becton-Dickenson Fibrometer) .....hrs.

2. Discuss Marketing with Sales
(Data: Note on Marketing) .....hrs.

E. Report to Client .....hrs.

Phase II. PRELIMINARY DESIGN

A. Preliminary Design Proposals .....hrs.

B. Discussion with Management, Productions, and Sales .....hrs.

C. Revised Design Proposal .....hrs.

D. Discussion with Management, Sales, and Lab Technicians .....hrs.

E. Design Proposal and Manufacturing Cost Estimate .....hrs.

F. Discussion, Approval on Approach to Design .....hrs.

Phase III. DESIGN DEVELOPMENT

A. Detailed Layout	.....hrs.
B. Hand Made Prototype	.....hrs.
C. Discussion with Management, Production, and Sales	.....hrs.
D. Final Corrected Prototype	.....hrs.

Phase IV. FINAL DESIGN

A. Production Drawings	.....hrs.
B. Follow Through with Suppliers of Component Parts	.....hrs.
C. Follow Through with Assembly of Final Product	.....hrs.
Total Time	.....hrs.
Fee for Design Services	$.....

B. Operations Analysis

An important problem area not suggested for solution by Mr. Sullivan during his initial discussions with Mr. McKim was the difficulty experienced in manipulating the timer switch, pipette, and clot hook simultaneously when the prothrombin time test was performed. The rubber tube and attached pipette hung awkwardly from the operator's mouth after the reaction was initiated because both hands were occupied. The client, of course, rarely wholly defines the problem for the designer; he does not possess the prerequisite knowledge. This case study has been structured to emphasize to the student that he is a "problem-finder" as well as a problem-solver by virtue of his design knowledge and experience.

One knowledgeable approach to the above mentioned human factors' problem is to conduct an operations analysis. If such an analysis is performed, it is suggested that the first step be to note each operation in sequence, which hand is used, and where the visual emphasis is at that moment. The following outline summarizes an observation made at the Oxford Laboratories on August 15, 1961:

OPERATION PROCEDURE - OXFORD LABORATORIES PROTHROMETER

OPERATION	INSTRUMENT	HAND	VISUAL
1. Pipette reagent	Pipette	Right	Reagent
2. Pipette blood sample	Pipette	Right	Blood
3. Simultaneously:			
a. Push timer switch	Timer switch	Left	Slide
b. Pipette blood and reagent to slide	Pipette	Right	Slide
c. Drop pipette, pick up clot hook; agitate solution	Clot Hook	Right	Solution
4. Simultaneously:			
a. Solution coagulates	Clot Hook	Right	Solution
b. Turn off timer	Timer switch	Left	Switch
5. Read timer counter	Timer counter	----	Counter read-out
6. Turn counter to zero	Counter dial	Left	Counter read-out

In the second part of the operations analysis, it is suggested that the student note the paths of motion of "links" between the Prothrometer, doctor, instruments, and patient, using a plan view or other diagram. A line on the diagram should be drawn to represent each link between the doctor's hands and the respective instrument for each operation in the test sequence. The test sequence can be simplifed by uncrossing and rearranging the links according to their importance, sequence, and frequency. Simultaneous visual links can be indicated on the same diagram, joining the points of visual interest. For more specific information on link analysis, consult the Human Engineering Guide to Equipment Design, McGraw-Hill, 1963, Chapter 8.

C. Special Design Problems

In addition to the design problems described above, the following problems were noted by Mr. McKim upon more intensive analysis of the Prothrometer:

1. The heat block is actuated by plugging into a wall socket. This feature requires the operator to unplug the instrument when it is not in use.

2. The glass slide is not positively positioned. The slide may upset if the Prothrometer is jarred or moved.

3. The thromboplastin is warmed prior to conducting the blood test by placing it in a precarious position on top of the heat block.

4. The instrument is unstable since it has only three feet.

5. The timer read-out is in a questionable location.

6. The externally visible components are not visually stressed in accordance with their functions.

7. The graphics are crude. It is necessary to consider the relative importance of "Oxford", "Prothrometer", "Timer", etc. The graphics should be considered at the onset as an important functional and visual part of the design and not applied as a final touch to a firm product configuration.

8. The toggle type timer switch is crudely unsuited to its function.

9. The heat indicator light location does not express what part of the instrument is operating.

10. The front face of the Prothrometer is not normal to an "average line of vision".

11. The blood and reagent solution should be linked to the heat reservoir with heat conductive material and a minimum air gap.

12. The Prothrombin Test is temperature sensitive and must be conducted at body temperature. Temperature stabilization is achieved at the Stanford-Palo Alto Hospital, for example, by immersing vials of thromboplastin reagent in a water-bath maintained at 37° C $\pm$ 1/2° C, which was sufficient accuracy for reproducible test results with a given blood sample. The thermostatic control of the Prothrometer is pre-set by Oxford to maintain the reagent bottle and blood-reagent mixture at body temperature when the instrument is used in a normally heated room. The heat block is maintained at 1° C above body temperature to compensate for heat losses by radiation and convection over the thermal path between heat block and glass slide. Experience has indicated that the pre-set thermostat rarely needs to be reset. The exposure of the thermostat reset screw may, therefore, invite uninformed adjustment.

13. The design of the Prothrometer does not separate the "medical look" from the "kitchen look" or the "automotive look". The Oxford Prothrometer will establish the "Oxford look". In the two years after the introduction of Mr. McKim's new design, Oxford has introduced two new products, and the

corporate identity of the Oxford line was established by the new Prothrometer design.

14. The selling price of the Prothrometer does not reflect the mark-up common to the medical instrumentation industry of four to five times the manufacturing cost. The selling price of the new instrument designed by Mr. McKim was $157.50.

In summary, it is suggested that these problems and discussion areas be discussed by the instructor if the students do not suggest them independently. Although the problem may appear to the student at first glance to be a "simple re-design", deeper study indicates technical, production, human factors, aesthetic factors, and marketing problems which truly tax the designer's ability.

NOTE ON MARKETING

The Oxford Prothrometer uses the "Whole Blood Micro-Method" to determine prothrombin time. U.S. medical laboratories, on the other hand, use the "Plasma Method". Therefore the Oxford Prothrometer is not primarily directed to the U.S. "medical laboratory market". It is directed to the "doctor market".

The "Whole Blood Micro-Method", which is the popular European laboratory technique, has two basic advantages over the "Plasma Method":

1) It is simpler and faster because the blood is not reduced to plasma before testing.

2) It is less painful to the patient because only a drop of blood is drawn from the finger. (Vein puncture is especially painful for patients with small, hard, or deep veins.)

These advantages make the Oxford Prothrometer concept appealing to doctors who would like to determine the patient's prothrombin time almost painlessly while the patient is in the office, and without resort to a medical laboratory.

The Oxford Prothrometer is used by the doctor or his nurse. The materials and labor for one test cost approximately 15 cents. The test takes approximately one minute. The doctor can normally charge the patient $5.00 for the determination. Therefore the doctor, by not resorting to the Plasma Test and a medical laboratory, has:

1) a happier patient (no needle in the vein)

2) an immediate basis for prescribing new anti-coagulant therapy

3) a tidy profit

An important part of Oxford's Prothrometer concept is the utilization of reagents and supplies which will be reordered from Oxford. In 1964, sales of reordered supplies constitute a significant portion of the Oxford income.

Oxford has, however, found that it is difficult to sell to doctors. Doctors, as a group, are extremely busy people. In addition to caring for their patients, they must keep up with new medical knowledge and techniques. Pharmaceutical houses literally innundate doctors with expensive literature and samples. Since many patients are loyal readers of the Reader's Digest, doctors must keep "one up" on the latest "miracle" pills or operations. It is difficult to obtain the cooperative attention, and thereby to sell, to the doctor market because the doctor's attention is constantly divided between patients, medical journals, and intensive pharmaceutical sales efforts.

Further, in the case of an instrument such as the Prothrometer, the doctor who buys the product often does not personally use it. His nurse uses it. Nurses, who not rarely believe that they are already overworked and underpaid, resent the addition of a new instrument to the office, or they use the new instrument as a wedge for a pay raise. In some offices, this interpersonal situation has firmly closed the door to new instruments.

How does Oxford sell to the difficult "doctor market"? Office-to-office selling, by an Oxford sales force, would be prohibitively expensive in relation to the price of the instrument. (Pharmaceutical houses can afford this kind of marketing because they promote many products, some of which are very expensive.) But a new instrument concept must be demonstrated to effect a sale. Therefore Oxford uses the following marketing methods:

1) It demonstrates the Prothrometer at medical meetings. Meetings of cardiologists or internists are especially fruitful. The doctor has more free time at a meeting and he is seeking new ideas.

2) It establishes regional dealerships. The dealer's salesmen, trained in the use of the Prothrometer, call "office-to-office". This kind of marketing is financially rewarding to the dealer who represents a number of manufacturers. Dealer marketing is most effective in the Southern states.

3) It advertises a "trial offer" in selected medical journals. Approximately 40% of sales are effected in this way - self demonstration - without a sales demonstration. (Visual clarity of function, therefore, is an important design objective.)

In addition, satisfied doctors have read technical papers at medical meetings which praise the Prothrometer. These, and "word of mouth" have also helped promote the use of the instrument within the "doctor market".

The Prothrometer will measure prothrombin time of plasma quite as well as of whole blood. Therefore the Prothrometer can also be used in the medical laboratory -- especially in the small lab where few determinations per day are required. Oxford Laboratories, in its product development subsequent to the Prothrometer, has concentrated on the medical laboratory market. Using its own two salesmen plus the national sales force of a large medical laboratory supply house, Oxford is currently selling Prothrometers to the medical laboratory as well as to the doctor market.

The medical laboratory, however, is rapidly becoming automated. Should Oxford direct a new prothrombin time device toward the lab market, it would certainly keep this trend well in mind. The current Oxford device, designed for doctors, fills an area of need within the medical laboratory market. But it was not designed for the medical laboratory

environment or for the intellectual and psychological characteristics of medical laboratory technicians. A suggested area of investigation: how should two prothrombin time devices - one directed to the doctor market, and one to the medical laboratory market, differ? This is a question which cannot be answered by the designer who speculates idly at the drafting board.

This marketing note, which includes information that was not available at the time the Prothrometer was redesigned, has been prepared in the hope that it will initiate a discussion about the market for, and marketing of, a new Oxford Prothrometer.

ECL-18
ME 116

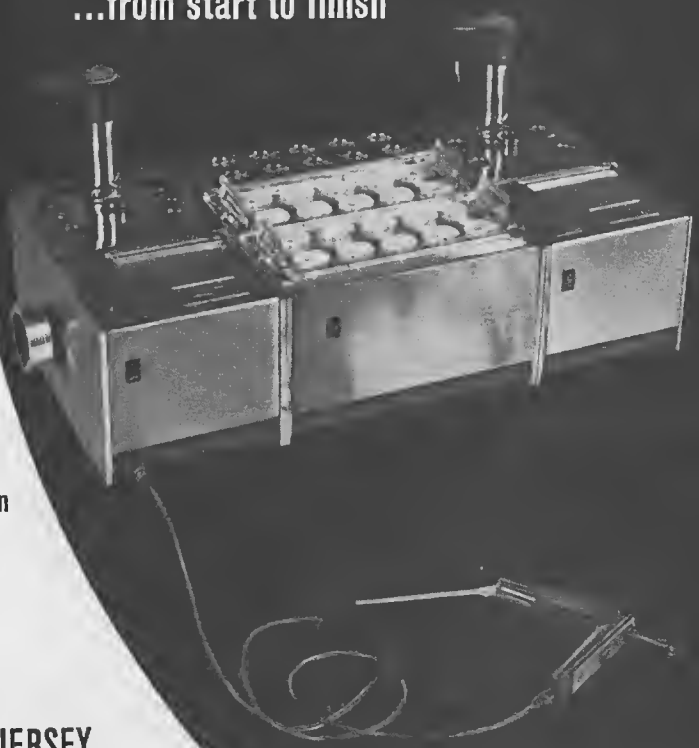
NOTE ON BECTON-DICKENSON FIBROMETER

8 components to "automate" your prothrombin time testing

VACUTAINER Specimen Collection Tubes
FIBROMETER Precision Coagulation Timer
FIBROMETER Automatic Pipette
FIBROMETER Automatic Pipette Disposable Plastic Tips
FIBROMETER Precision Heating Unit
FIBROTUBE Disposable Coagulation Tubes
CALSOPLASTIN Thromboplastin Extract
Normal Control Sera

new FIBROMETER™ system

...from start to finish



The FIBROMETER Precision Coagulation Timer is an electromechanical instrument for measuring plasma coagulation time automatically, objectively, and reproducibly. It duplicates skilled manual testing but saves time and avoids inaccuracy induced by fatigue. As part of a modular system, one or more FIBROMETER units may be joined together. A Thermal Prep Block Precision Heating Unit preheats plasma and reagents to 37°C., a FIBROMETER Automatic Pipette dispenses plasma and reagents uniformly, and starts the timing cycle automatically. Clot formation end point is sensed and recorded electronically and results are read on a Digital Readout counter. The combined FIBROMETER System fills a decided need for completely automating prothrombin time determinations for diagnosis and anticoagulant therapy control.

Products of B-D LABORATORIES are available through your local distributor.

B-D

B-D LABORATORIES, INC., RUTHERFORD, NEW JERSEY

B-B-L Baltimore Biological Laboratory

Falcon Plastics

Science Education Products

In Canada: Becton, Dickinson, & Co., Canada, Ltd., Toronto 10, Ontario
Overseas: Becton, Dickinson & Co., S.A., P.O. Box 1173, Colon, Free Zone, Panama

Courtesy of Becton-Dickenson Laboratories

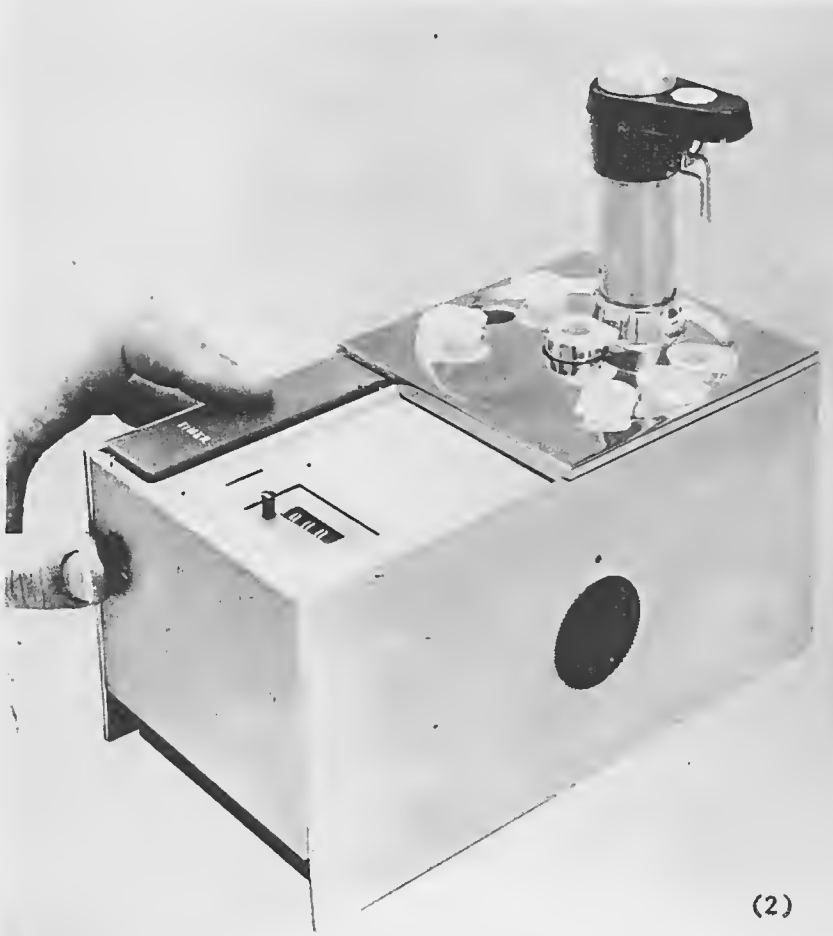
Products to "Automate" Prothrombin-time Testing

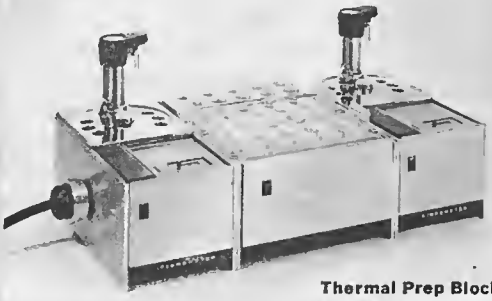
For prothrombin-time determinations, this new, mechanically accurate, time-saving system elevates the test beyond human calculation . . .

Your technologists can run plasma end-points on a FIBROMETER Precision Coagulation Timer without learning any new, involved methods. This compact, miniaturized machine utilizes traditional manual techniques to record end points accurately, automatically and reproducibly.

You can handle any size workload with the FIBROMETER system. Large-capacity Thermal Prep Blocks are available separately, so is a fast, accurate Automatic Pipet, so are the specially designed small, disposable FIBROTUBE units that hold plasma and reagents. Even the thromboplastin extracts you need are available from B-B-L—choose either new CHROMOPLASTIN chromatically labeled thromboplastin extract or CALSOPLASTIN non-labeled thromboplastin extract.

It took technical know-how and technical resources to put this complete prothrombin-time system where it belongs—at your fingertips! B-B-L is the only manufacturer supplying both completely interstandardized reagents and apparatus for prothrombin time determinations. Ask us for literature—or a demonstration.

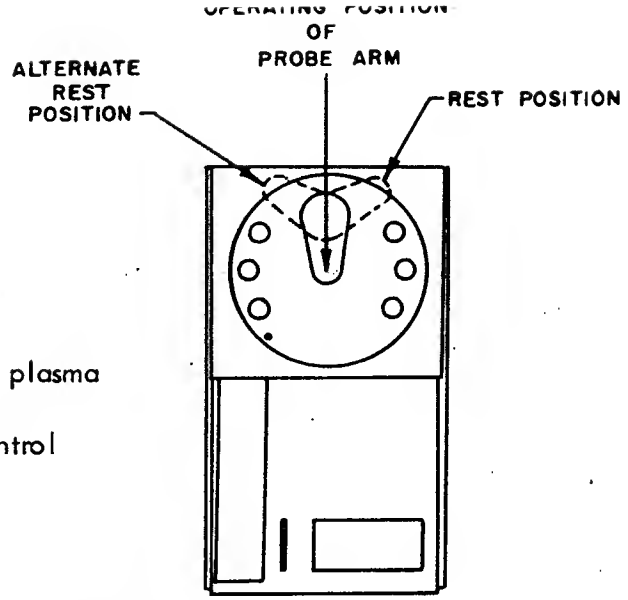




Thermal Prep Block
available shortly

Automatic Pipet
available shortly

BECTON-DICKENSON FIBROMETER, CON'T

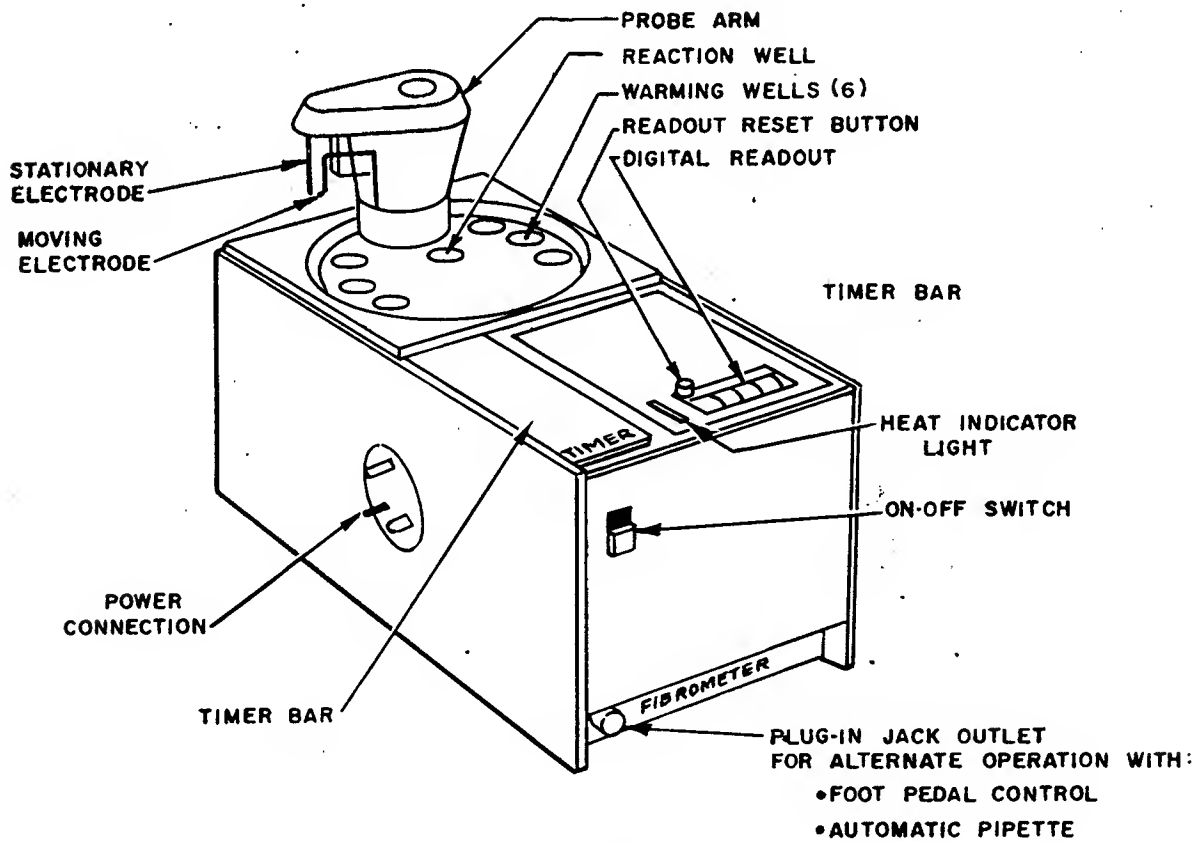


FIBROMETER
Precision Coagulation Timer

An electro-mechanical instrument

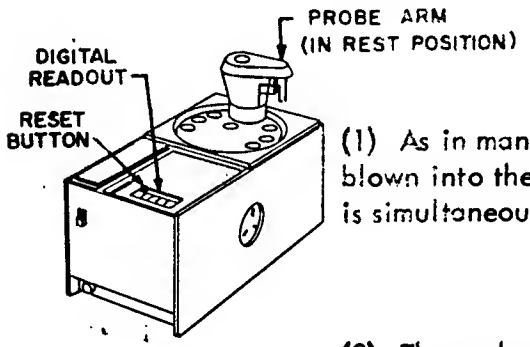
to measure coagulation properties of plasma

for diagnostic use and in therapy control



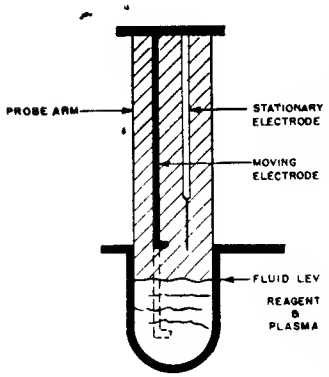
BECTON-DICKENSON FIBROMETER, CON'T

The basic action of FIBROMETER, Precision Coagulation Timer, is quite simple. It duplicates a time-tested manual technique. Note that the electrode action is identical to that of the trained technician using a wire loop. Guesswork in determing the end-point is eliminated.

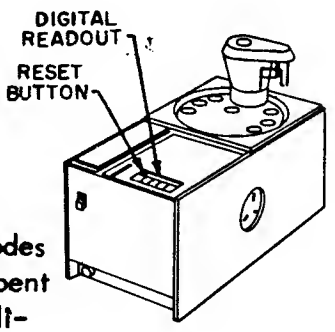


(1) As in manual techniques, aliquots of plasma are blown into the thromboplastin reagent; the timer bar is simultaneously pressed to initiate mechanical action.

(2) The probe arm automatically swings into position over the reaction well and drops into place. The moving electrode alternately descends and lifts in a sweeping motion, to seek and sense initial clot formation.



(3) When the end-point occurs, the electrode and the timer stop. Prothrombin time, in seconds and tenths, is registered on the digital readout.



(4) The readout reset button is pressed; the electrodes are cleaned by wiping with laboratory grade absorbent tissue and the probe is repositioned at rest, in readiness for the subsequent test.

BECTON-DICKENSON FIBROMETER, CON'T

THE DETECTION-SENSING ACTION

Formation of the insoluble fibrin network in the reaction mixture serves to complete the electrical circuit through the detection-sensing system, thus triggering an electronic mechanism which amplifies the signal to stop the timer. The circuitry is transistorized for long, rugged, maintenance-free operation.

THE PROBE UNIT

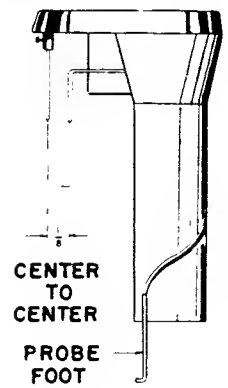
Following activation of the timer, a built-in delay of approximately 1.5 seconds occurs after which the probe arm automatically swings into position over the reaction well and drops into place. This delay permits unhampered access to the reaction well during pipetting and facilitates accurate, unhurried pipetting. The delay in nowise affects test accuracy.

Each probe unit may be used interchangeably with any FIBROMETER.

THE ELECTRODES

The stainless steel from which the electrodes are constructed is a grade specific for biological use. It has high flexural strength.

One electrode remains stationary within the reaction mixture. The other, a moving electrode, is cycled through the mixture every half second. This provides exceptional resolution power.



Exact spacing of the moving and stationary electrodes is not critical, although gross distortion should be avoided. Should the wires become bent through by accident, the sketch at left will serve as a guide to bending them back into proper shape.

The probe foot is critical to accuracy. Although its exceptionally heavy construction makes damage unlikely, should this occur, return of the complete probe arm for factory adjustment is recommended.

NOTE ON COMPONENT REFERENCES

Technical data, prices and other information on switches, timers, thermostats, heating elements, and other components may be obtained from the following sources:

Switches, Miscellaneous Components

1. Electronic Engineers Master Catalog
   Tech Publishers, Inc.
   645 Stewart Avenue
   Garden City, New York

2. Sweet's Product Design Catalog
   Sweet's Catalog Service
   F. W. Dodge Corporation
   119 West 40th Street
   New York 18, New York

Timers, Counters

1. Precision Timer Company, Inc.
   66 Coulter Street
   Saybrook, Connecticut

2. Cramer Controls Corporation
   Miller Street
   Centerbrook, Connecticut

3. Veeder-Root, Inc.
   Hartford 2, Connecticut

Thermostats

1. Vulcan Electric Company
   Danvers, Massachusetts

2. Fenwal, Inc.
   Ashland, Massachusetts

3. Stevens Manufacturing Co., Inc.
   Mansfield, Ohio

4. Burling Instrument Company
   16 River Road
   Chatham, New Jersey

Heating Elements

1. Vulcan Electric Company
   Danvers, Massachusetts

2. Trent, Inc.
   201 Leverington Avenue
   Philadelphia 27, Pennsylvania

3. General Electric, Inc.
   Industrial Heating Department
   Shelbyville, Indiana

4. Chemelex, Inc.
   Mineola, New York

NOTE ON MANUFACTURING COSTS

The following figures give the manufacturing costs of the existing Oxford Prothrometer:

Item	Manufacturing Cost[1]
Sheet Metal Case	$ 5.37
Painting	1.37
Heat Block	6.00
Timer Assembly	7.00
Heating Pad	2.75
Thermostat	6.50
Toggle Switch	0.25
Pilot Light	0.30
Power Cord	0.18
Decal	0.15
Internal Wiring	0.20
Miscellaneous Parts	0.10
Sub Total:	30.17
Assembly, 2 hours at $6.00 per hour	12.00
Overhead, Engineering	8.00
Total Manufacturing Cost:	50.17

The accessories used with the Prothrometer were provided at additional cost to the customer. These included a carrying case, four pipettes, two thermometers, two clot hooks, two dozen lancettes, two dozen spot bandages, one bottle each for alcohol, ketone, and distilled water. These items cost Oxford Laboratories $7.50 per accessory set in lots of 500 sets.

1. Per-item cost when purchased in lots of 500 items.

NOTE ON ASSEMBLY DIFFICULTIES

The following photographs were taken during the assembly of the Prothrometer:

Figure 1. Prothrometer Components

Figure 2. Attaching Motor to Bracket

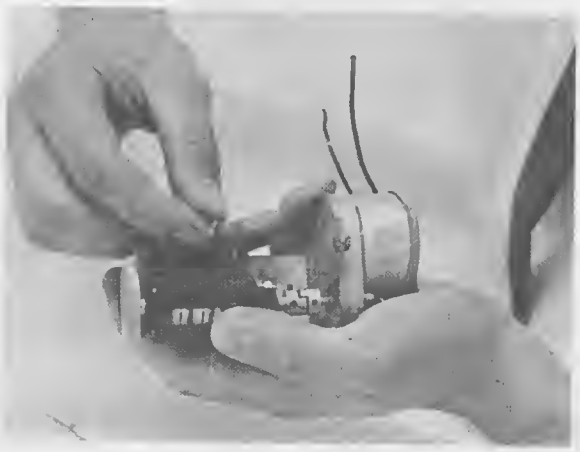


Figure 3. Attaching Counter to Bracket

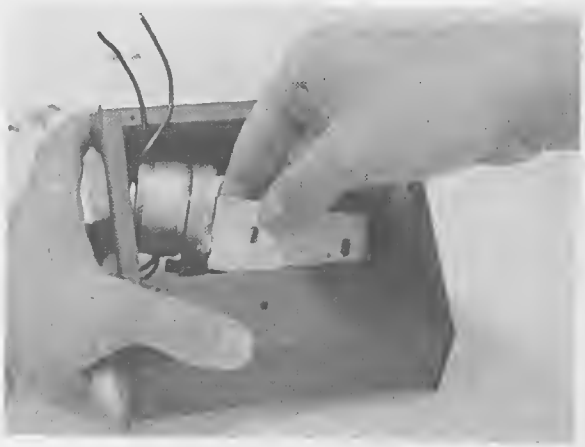


Figure 4. Inserting Timer Assembly Into Sheet Metal Case

ASSEMBLY DIFFICULTIES, CON'T

Figure 5. Securing Timer Assembly

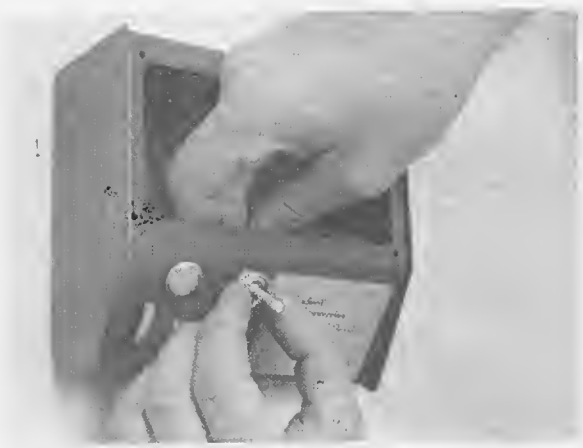


Figure 6. Installing Toggle Switch

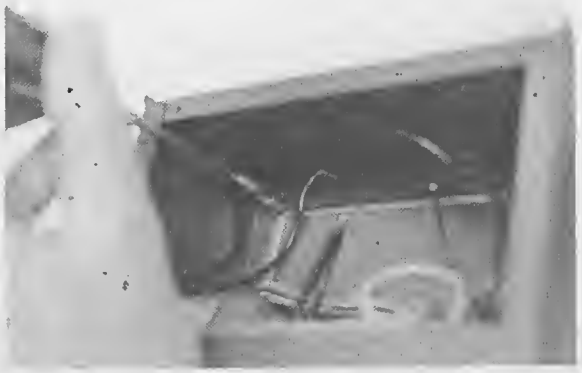


Figure 7. Attaching Heat Block

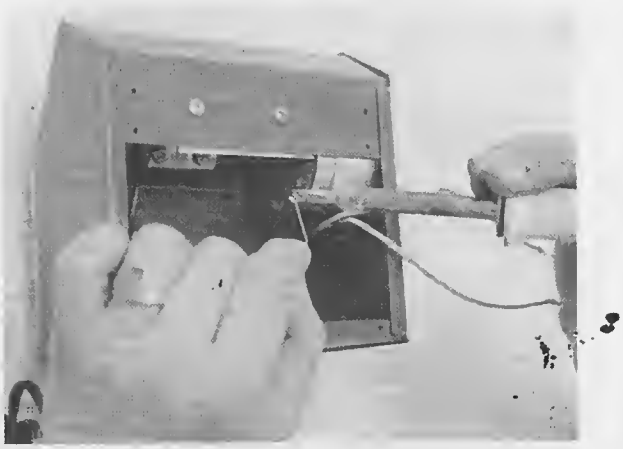


Figure 8. Soldering Electrical Leads

NOTE ON THE BLOOD TEST SEQUENCE

The following photographs were taken during a Prothrombin Time Test performed at the Stanford Medical Clinic:

Figure 1. Patient and Intern Seated in a Clinic Office

Figure 2. Lancing the Patient's Finger

Figure 3. Pipetting Blood

Figure 4. Pipetting Reagent

Figure 5. Ejecting Blood and Reagent

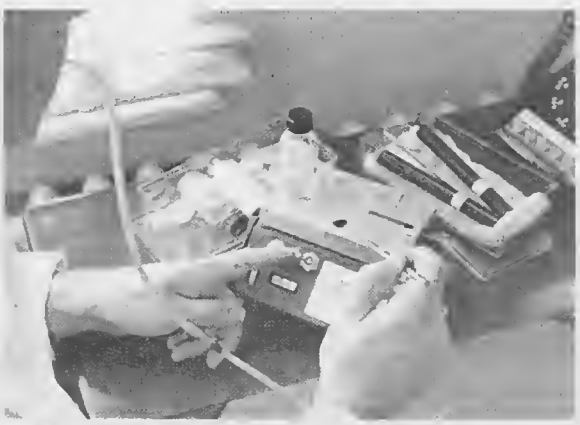


Figure 6. Stirring the Mixture

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Refer to: Glover, R. P., and Kuzell, W. C.: Prothrombin time—determination by a whole blood micromethod for control of anticoagulant therapy, Calif. Med., 95:24-29, July 1961.

Prothrombin Time

Determination by a Whole Blood Micro-Method for Control of Anticoagulant Therapy

RICHARD P. GLOVER, *M.S.*, and WILLIAM C. KUZELL, *M.D.*, San Francisco

A micro technique that is here described for "prothrombin time" determinations, employing capillary whole blood, provides a range of values which is closely correlated with the Quick one-stage plasma method, thus providing interchangeability of results both in normal persons and in patients who have been treated with anticoagulant drugs.

Avoidance of the use of a water bath and centrifuge permit this technique to yield immediate results at the bedside, in the office or in the patient's home.

The use of a whole blood instead of a plasma technique lends additional safety to control of anticoagulant medication, since it may reflect depression of clotting factors not apparent by the usual plasma methods.

PROPER USE of oral anticoagulants demands reliable laboratory control.[4] We shall describe a simple micro-method for testing whole blood "prothrombin time" or "activated clotting time." This method is applicable to clinical control of anticoagulant medication.

Ideally, laboratory tests to control anticoagulant medication should be devised so that:

1. *In vitro* conditions in the performance of the test should approximate as nearly as possible the *in vivo* conditions.

2. Results of any method should be readily reproducible and permit favorable comparison with other standard one-stage methods, in both absolute and relative aspects.

3. Only micro-quantities of capillary blood should be employed, thus avoiding the frequent venipuncture which may be required on a single patient.

4. Results should be available immediately for the guidance of the physician in regulation of anticoagulant dosage.

5. The interval between drawing blood and testing should be as short as possible to avoid erroneous results due to adverse effects of storage.

6. Performance of the test should be technically simple so that, in addition to laboratory personnel, physicians, nurses, or sometimes even patients themselves may achieve proficiency in the determination.

7. Employment of cumbersome equipment such as centrifuges and water baths should be eliminated so that tests may be done at bedside, in the patient's home or in the office of the physician responsible for anticoagulation administration.

Usually for the control of oral anticoagulant therapy as previously practiced, a plasma method is used, the end point of which is determined by recalcification of plasma in the presence of added thromboplastin. This is commonly designated as the one-stage "prothrombin time" or the Quick method.[14] As the therapeutic modality of anticoagulation has become more extensively employed, certain disadvantages, both practical and theoretical, have appeared in this time-honored technique. Among the disadvantages of the standard one-stage Quick method are:

1. Venipuncture is required. Usually 3 to 5 ml. of blood is taken.

2. Time for performance of this test, as usually dictated by custom in hospitals and clinical laboratories, is frequently as much as several hours after venipuncture. This time lag greatly lessens the usefulness of the test to the clinician, involving additional communication with the patient or the nurse.

3. Errors arise due to the effects of storage of citrated or oxalated blood. These sources of error include alteration of contact factor (Hageman factor),[9,11,19] labile factor (Factor V),[1,2] platelet Factor I, antihemophilic globulin (Factor VIII),[11] and antithrombin.[2]

4. Performance of the plasma test requires a centrifuge and a water bath, restricting its use to laboratories which often do not function during the entire 24 hours of the day.

5. There is considerable variability in the results from one laboratory to another, both with respect to normal and therapeutic ranges.[10]

6. Tests employing plasma have the theoretical disadvantage of being insensitive to possible deficiency of Factor IX (Christmas Factor, Antihemophilic Globulin, PTC).[9,12]

From the Arthritis Clinic, Presbyterian Medical Center, San Francisco 15.

Submitted March 22, 1961.

The clinical need for a test giving immediately available results has led to various methods of employing mixtures of whole blood and thromboplastin to determine "activated clotting time" rather than plasma "prothrombin time."*

Whole blood methods that neglect control of critical temperatures have not yielded results comparable to those obtained by plasma methods in absolute terms of seconds elapsed. Other methods in which temperature control is included, such as one reported recently by Phillips and coworkers,[15] while giving excellent relative correlation with the Quick plasma method, fail to correspond in absolute values, especially in the higher ranges of anticoagulation. Such disparity demands radical revision of the clinician's concept of a safe therapeutic range, making for confusion when compared with standard Quick determinations.

Hoffman and Custer[7] in 1942 reported a micromethod at 37° C. for determination of "prothrombin time" on fresh capillary blood. Since physical conditions were controlled as in the original Quick procedure, their values were comparable both in absolute and in relative terms. They observed that variation in hematocrit caused no divergence in comparing plasma with capillary whole blood techniques. By dilution techniques their micro-method showed ". . . a parallelism with the Quick values usually within 5 per cent prothrombin."

The essential requirements of accurate temperature control, elimination of water bath and centrifuge, use of fingertip blood, and convenient portability seem to have been satisfied in the method herein reported.

MATERIALS AND METHOD

Instrument

A portable instrument‡ (14x15x9.5 cm.) provides a constant temperature for performance of the test. A built-in timing device composed of a small synchronous motor and a counter calibrated to 0.1 seconds is used to measure the observed clotting time.

A soft glass capillary tube (1.2-1.4x75 mm.) is employed to draw up the blood and the thromboplastin. A piece of soft rubber tubing, as used for standard blood-diluting pipettes, is equipped with a plastic mouthpiece at each end. A small rubber bulb similar to those used with smallpox vaccine tubes is stretched over the end of one of the plastic mouthpieces. The capillary tube is inserted into the other end of the vaccine bulb.

The clotting reaction takes place in the dimple of a glass depression slide which rests on a heated aluminum block, the temperature of which is thermostatically controlled at 38.5° C. Measurement of the surface temperature of the glass slide varies from 37° to 38° C., depending upon the ambient temperature and convection currents in the air.

Thromboplastin

Commercially available rabbit brain-lung thromboplastin† was employed. When using capillary blood the presence or absence of added calcium ions in the commercial thromboplastin mixture is immaterial, since the patient's own whole blood calcium is sufficient for the clotting action *in vitro*. If, for some reason, oxalated or citrated venous blood is tested, the thromboplastin mixture must contain added calcium chloride.

Method

1. After both the instrument and the thromboplastin reagent have attained the optimum temperature of 37° C., the finger or ear lobe is punctured cleanly and deeply enough to insure a free flow of blood without excessive squeezing. Whole venous blood may be used, provided the optimal ratio of citrate or oxalate is employed.

2. The first drop of blood is wiped away and the fingertip or ear lobe is gently squeezed to produce a large fresh drop of capillary blood.

3. A soft glass capillary tube is employed to draw up the blood to about one third of its capacity. The tip of the capillary tube is then wiped clean to avoid contamination of the thromboplastin reagent, and the blood is drawn 2 or 3 mm. farther up into the tube. This aids in preventing premature mixing of the blood and thromboplastin within the tube.

4. Without delay, an amount of thromboplastin approximately equal to the volume of blood in the tube is drawn up and the contents of the tube are then expelled into the dimple of the warmed glass depression slide. The timer is started.

5. With a clean metal "clot hook" (a scleral retractor is suitable), the blood-thromboplastin mixture is stirred in a rapid rotary fashion. The timer is stopped when gross clotting is observed. With normal blood and also with blood from patients receiving anticoagulants the entire mixture appears to clot simultaneously.

PRECAUTIONS

Accuracy and close agreement of results are dependent on absolute adherence to obvious, but easily overlooked, manipulations. Among these are:

*References 7, 8, 15, 16, 17, 18.

‡Prothrometer® manufactured by Oxford Laboratories, Redwood City, California.

†Simplastin® manufactured by Warner-Chilcott Co., Morris Plains, New Jersey.

1. Enough time should be allowed for the heating block of the instrument and the thromboplastin reagent to attain optimum temperature before proceeding with the test—usually about 10 to 15 minutes if the machine has been at room temperature.

2. The thromboplastin reagent should be fresh and should not have been contaminated by blood from previous determinations. When possible, the consistent use of a single commercial source of thromboplastin helps to insure uniformity of results.

3. Free-flow of capillary blood is essential. Excessive squeezing of the finger or earlobe will introduce tissue fluids which may alter the test. If more than two determinations are to be made at one time, another area should be pricked.

4. The ratio of the capillary blood and the thromboplastin reagent should be close to 1:1. Premarking of the capillary tubes is helpful in this measurement.

5. Blood should be drawn into the capillary tube before the thromboplastin. Reversal of this sequence increases the likelihood of premature mixing of the reactants due to differences in their relative viscosities.

6. In doing duplicate determinations, time may be saved by not resetting the timer and merely making a mental note of the initial determination.

RESULTS

Using the procedure outlined above, the "prothrombin time" or "activated clotting time" on probable normals lay in the range of 10 to 13 seconds, thus comparing favorably with results of the Quick method. It has been our practice to report results in seconds rather than as a "per cent of normal." In order to establish the viability of the thromboplastin, a "normal" was determined by random sampling or by utilizing commercial pooled plasma. Thromboplastins that yielded a "normal" value greater than 14 seconds were not used.

In establishing a comparison of the whole-blood microcapillary method with the standard one-stage plasma test of prothrombin time, every effort was made to maintain ideal test conditions. Dilution of venous blood and citrate was made with volumetric precision, employing graduated centrifuge tubes. When difficulty with venipuncture necessitated several attempts, the samples were discarded. The citrated venous blood was centrifuged immediately for 5 minutes at 1,600 r.p.m. and the plasma layer removed at once. In all instances the macro-method determination was performed within 15 to 20 minutes following venipuncture. When the micro-technique was applied both to finger-puncture capillary blood and to freshly drawn citrated venous blood, close agreement of results prevailed. Progressive shortening of capillary blood prothrombin time was noted following second or third determinations obtained from a single puncture site, thus underscoring the necessity of fresh, free-flowing capillary blood for accurate analysis.

The same vial of lyophilized thromboplastin was used for both methods in each comparative determination, and it was not used more than 48 hours after reconstitution even though it was preserved at 5° C.

The data that were analyzed consisted of 205 pairs of observations obtained by the whole blood micro-method and the plasma macro-method (Quick), divided for statistical purposes into groups "A" and "B" consisting of 182 and 23 pairs respectively.

In group "A" (182 pairs) each observation on the micro- or macro-methods was the average of two determinations. Group "B" (23 pairs) included 20 pairs of observations from single, duplicate or triplicate determinations and 3 pairs where the whole blood micro-method gave an unknown reading greater than 50 seconds. All comparative results are combined on the scattergram shown in Chart 1.

For group "A" (182 pairs) the estimated regression line is

$$y = .08 + .96x,$$

where x and y represent the prothrombin times for the micro- and macro-methods, respectively, and the correlation coefficient (r) between x and y is $r = +.96$. The statistical analysis* established a definite linear relationship between the methods, even though it is not the ideal direct relationship $y = x$. For prothrombin times up to 50 seconds, the estimated regression line lies significantly (at the 1 per cent level) below the line $y = x$; that is, the micro-method tends to give a slightly higher prothrombin time than the macro-method. However, at the point of maximum difference, namely, at a prothrombin time of 50 seconds, the average statistical discrepancy between the two methods is just 2 seconds.

In calculating the statistical correlation, three instances involving two patients were omitted. In these three instances the microcapillary method produced results much longer than the macroplasma method. The macro-method showed values of 33.0, 20.6, and 39.6 seconds, as compared with microcapillary figures in excess of 50 seconds. We omitted these three comparative tests because prolongation of micro-test results apparently reflected the diminution of coagulation factors other than those in the *extrinsic* coagulation system. It is generally agreed that drugs of the coumadin type act to depress both the *intrin-*

*By Dr. Rupert G. Miller, Jr., Departments of Statistics and Preventive Medicine, Stanford University.

PROTHROMBIN TIME TEST CON'T

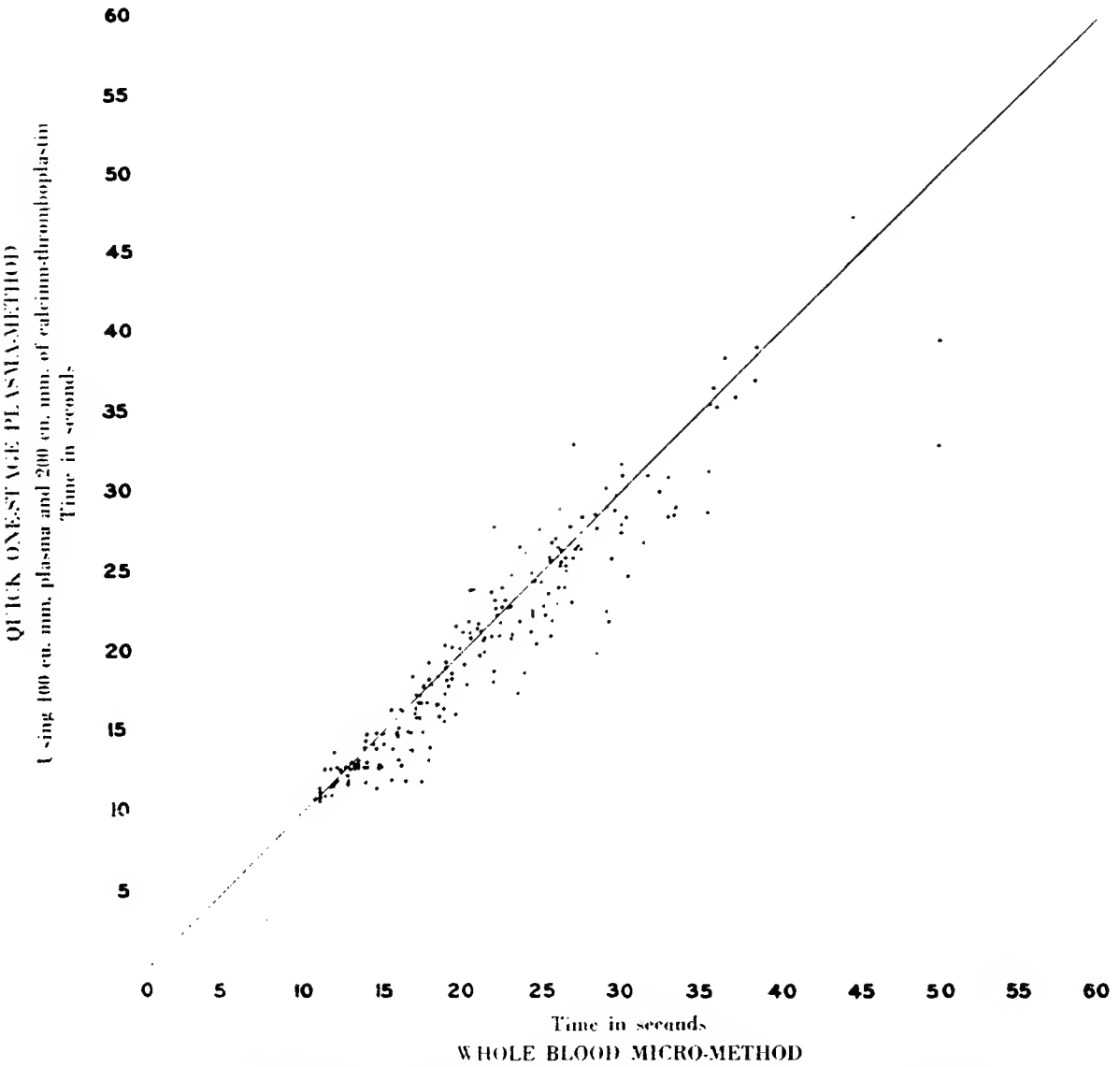

Chart 1.—Comparison of "prothrombin time" determinations by the Quick one-stage plasma method and the whole blood micro-method.

sic and the *extrinsic* clotting systems.† Prothrombin (Factor II) and Stuart-Prower factor (Factor X) are affected in both systems, as is Christmas factor (Factor IX) in the *intrinsic* system and proconvertin or stable factor (Factor VII) in the *extrinsic* system.[12] The one-stage plasma (Quick) test is not sensitive to depression of Factor IX but reflects alteration of activated clotting time associated only with the *extrinsic* system.

Two of the three test pairs showing longer micro-capillary than macroplasma prothrombin times occurred in one subject, a 79-year-old man with hepatic cirrhosis and gross hematuria. While the plasma macro-method gave figures (33.0 and 20.6 seconds) in the "therapeutic range of anticoagulation" his blood failed to clot at 50 seconds by the micro-capillary method. The third widely divergent comparison occurred in a 69-year-old arteriosclerotic diabetic patient who had pronounced gingival bleeding while his macroplasma prothrombin time was 39.6 seconds and clotting did not occur at 50

†Clotting factors present in circulating blood are designated *intrinsic*. Activation of the *extrinsic* system requires tissue thromboplastin. In the formation of thrombin by the *intrinsic* system the following factors are required: Factor II (prothrombin), Factor IV (calcium), Factor V (proaccelerin), Factor VIII (antihemophilic globulin), Factor IX (Christmas factor, plasma thromboplastin component, or antihemophilic B factor), Factor X (Stuart-Prower factor), Hageman factor, platelets ("cephalin"), and plasma thromboplastin antecedent. The *extrinsic* system requires Factors II, IV, V, VII, and X plus Factor III (tissue thromboplastin). Oral anticoagulants depress Factors II, VII, IX, and X.[12]

seconds by the microcapillary method. The discrepancies in these three instances cannot be explained by technical error. The presence of spontaneous bleeding while the "prothrombin time" done by the Quick method was in the "therapeutic" range would indicate that there was a diminution in a coagulation factor to which the conventional plasma method was insensitive. Exact delineation of the additional treatment-induced clotting defect would have required more selective methods—the thromboplastin generation test, for example.

DISCUSSION

In the use of anticoagulant therapy, patients with defects in accessory clotting factors occasionally will be encountered. Neither the technique described in this paper nor the one-stage plasma "prothrombin time" may be considered a diagnostic tool in such problems. These one-stage tests measure only the ability of blood to clot within a given time when exposed to excess tissue thromboplastin and calcium ion. Prothrombin may not be specifically assayed by one-stage techniques. A more specific assay of prothrombin concentration is possible by two-stage techniques or by hydrolysis of a selective substrate, as in the TAMe[6] method. Results are still expressed, however, in terms of clotting activity rather than a quantitative absolute value; furthermore, the technical difficulty involved in the more elaborate tests makes reproducibility troublesome, from one laboratory to another. Apart from its technical difficulty the TAMe procedure is unsuited to routine control of anticoagulant medication, since it does not reflect depression of other blood clotting factors which are influenced by oral anticoagulants.

The one-stage "prothrombin" test, whether performed on plasma or whole blood, does not reveal defects in thromboplastin formation. Additionally, the blood of a hypofibrinogenemic patient will have a prolonged one-stage "prothrombin time" in the presence of normal plasma concentration of prothrombin.[2] In spite of these limitations, the one-stage tests are clinically accepted as guides to oral anticoagulant dosage. There are, however, three areas in which a rapid method for measuring "prothrombin" time of capillary blood appears to offer a distinct increase in safety and accuracy over the conventional plasma test. These situations involve the elimination of innate sources of error associated with: (1) Storage of plasma or blood, (2) repeated venipuncture, (3) disturbance of critical calcium ion concentration essential for accuracy in the one-stage plasma technique.

1. Since the microcapillary whole blood test is completed within seconds after initiating bleeding, the possibility of introducing additional coagulation factors, either acceleratory or inhibitory, due to storage of the blood, is largely eliminated. It is not uncommon for at least a half to three-quarters of an hour to elapse between venipuncture and centrifugation of the specimen. Frequently as much as several additional hours of storage intervene before the daily determination of "prothrombin times" is completed in a given laboratory. Delays in testing are owing in part to the convenience of waiting for a backlog of tests to accumulate so that a number can be done at one time, and partly to lack of cognizance by laboratory personnel of the various factors that may drastically alter the one-stage test. Chief among these storing phenomena are:

(a) *Labile Factor* (Factor V, proaccelerin) is essential for the conversion of prothrombin to thrombin, and its diminution appears to be solely a consequence of storage.[2] Storage does not impair the actual prothrombin concentration, but the one-stage "prothrombin time" progressively lengthens as the plasma ages. Thus, in anticoagulated patients, the unrecognized diminution of labile factor may lead to incorrect interpretation on the part of the clinician of the degree of anticoagulation achieved.

(b) Acceleration of clotting time, as measured by one-stage methods, becomes apparent within two hours, due to presence of an "activation substance" produced by interaction between Hageman factor (contact factor) and plasma thromboplastin antecedent.[12] Hageman factor also serves to activate Factor VII in the *extrinsic* clotting system, even in the absence of calcium[19] and at refrigeration temperatures,[11] so that one may assume its influence to be undiminished in refrigerated plasma. Comparative trials with plain glass and siliconized capillary tubes in the whole blood micro-method indicated that the period of contact between glass and blood is so brief as to be of negligible concern in this technique.

To avoid the influence of contact factor when glass tubes are used for collection of venous blood, Owren stressed that normal blood should be tested within a few minutes and blood from anticoagulated patients within one hour after venipuncture.[11] Assurance of such prompt disposition of the procedure cannot be gained from most laboratories doing the plasma one-stage technique.

(c) *Platelet Factor I* (platelet accelerator) acts to accelerate the conversion of prothrombin to thrombin in stored plasma, and may result in spurious shortening of the "prothrombin time."[5]

2. In dispensing with the necessity for large amounts of venous blood, one removes the possibility of partial undetected coagulation which may commence in the syringe when venipuncture is difficult. Serum produced by this premature coagulation elicits an accelerator substance which promotes the conversion of prothrombin to thrombin during

the test procedure,[2] again making an erroneously short "prothrombin" time.

3. Critical control of calcium ion concentration demanded by usual plasma "prothrombin time" methods is not a factor in the capillary blood technique where the physiological concentration of calcium remains undisturhed. The artificial situation in which blood calcium is removed by oxalate or citrate during venipuncture and then replaced when the "prothrombin time" is determined lends itself to technical errors which would profoundly affect the result. Excess *in vitro* anticoagulant, which may be present when the ratio of blood to balanced oxalate is less than 9:1, continues to precipitate the calcium ion added during the prothrombin testing procedure and makes for an excessively prolonged "prothrombin time," and sometimes a clot does not appear at all. The existence of excess oxalate may also be encountered when the hematocrit is higher than normal, requiring an increased concentration of calcium ion in the test procedure. In the microcapillary technique with whole blood, it appears that only an excess of calcium ion is required, since little difference in results was noted whether the material used was a commercial thromboplastin combined with calcium chloride* or a product in which the calcium was supplied separately and could be used as thromboplastin alone.†

450 Sutter Street, San Francisco 8 (Kuzell).

REFERENCES

1. Alexander, B., and deVries, A.: Human prothrombin; quantitative studies on plasma labile factor and restorative effects of normal, hypofibrinogenemic and hemophilia plasma on prothrombin of stored plasma, J. Clin. Invest., 28:24, 1949.

2. Alexander, B., deVries, A., and Goldstein, R.: Prothrombin: A critique of methods for its determination and their clinical significance, N.E.J.M., 240:403, 1949.

3. Bailey, N. T. J.: Statistical Methods in Biology, Wiley, New York, 1959.

4. Editorial. Anticoagulants: A cooperative effort, J.A.M.A., 172:1484, 1959.

5. Fell, C., and Seegers, W. H.: Platelet factor 1 related to prothrombin activation, Can. J. Biochem. Physiol., 36:645, 1958.

6. Glueck, H. I., Sherry, S., and Troll, W.: Assay of plasma prothrombin with a synthetic substrate, Proc. Soc. Exper. Biol. and Med., 87:646, 1954.

7. Hoffman, O. D., and Custer, R. P.: A micro-method for determining prothrombin time on fresh capillary blood using standard physical conditions, Am. J. Med. Sci., 204:1-11, 1942.

8. Manchester, B.: The value of continuous (1 to 10 years) long-term anticoagulation therapy, Ann. Int. Med., 47:1202, 1957.

9. Matthews, J. M., and Walker, W.: Control of anticoagulant therapy—A trial of thrombotest, Lancet, 1159, 1959.

10. Messinger, W., and Weiner, M.: An evaluation of some clotting tests in patients receiving coumadin drugs, A.M.A. Arch. Int. Med., 105:727, 1960.

11. Owren, P. A.: The laboratory control of anticoagulant therapy in large groups of patients, Farmakoterapi, 15:92, 1959.

12. Owren, P. A.: Thrombotest—A new method for controlling anticoagulant therapy, Lancet, 754, 1959.

13. Phillips, G. E., Lupdecke, H. F., Breuchaud, J. S., and Lenahan, J. G.: A micro-method for the determination of prothrombin activity, J. Lab. Clin. Med., 56:659, 1960.

14. Quick, A. J.: The clinical application of the hippuric acid and the prothrombin tests, Am. J. Clin. Path., 10:222, 1940.

15. Reich, C.: The feasibility of using finger blood for prothrombin determinations, Am. J. Clin. Path., 30:19, 1958.

16. Schwager, P. G., and Jaques, L. B.: A simplified technique for the determination of prothrombin times, Can. M. A. J., 60:258, 1949.

17. Smith, H. P., Ziffren, S. E., Owen, C. A., and Hoffman, G. R.: Clinical and experimental studies on vitamin K, J.A.M.A., 111:380, 1939.

18. Thompson, B. A.: A modified Quick's one-stage test for the control of out-patients on anticoagulant therapy, J. Clin. Path., 13:176, 1960.

19. Waaler, B. A.: Contact activation in the intrinsic blood system; studies on a plasma product formed on contact with glass and similar surfaces, Scand. J. Clin. Lab. Invest., 11(supp. 37):133, 1959.

*Simplastin, Warner-Chilcott Co., Morris Plains, New Jersey.
†Permaplastin, C. W. Alban & Co., St. Louis, Missouri.

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NOTE ON ANTICOAGULANT THERAPY

Prepared by the Research Staff of Oxford Labs for distribution to Sales Personnel.

I. INTRODUCTION

Diseases of the blood's clotting (or coagulation) mechanisms attack over a million people each year in the United States. More than four hundred thousand persons die of such diseases annually. Most prominent amont these diseases are those that affect the heart ("heart attacks", or in medical terms - "myocardial infarction") and those that affect the brain ("strokes" or in medical terms - "cerebral thromboses").

A revolutionary advance has occurred in the treatment of these diseases in recent years. Several drugs, the anticoagulants, have been developed to increase the tendency of the blood to coagulate (or clot). Scores of research studies have substantiated the effectiveness of these anticoagulants. As a result, there has been a tremendous increase in the use of these drugs.

A patient who is receiving anticoagulant drugs must have his blood tested at frequent intervals. This test determines the length of time required for the blood to clot under standardized conditions. The resultant measurement is known as prothrombin time. The Prothrometer(R) is an instrument which makes possible the use of easily performed micro methods for conducting the prothrombin time test.

II. BRIEF OUTLINE OF TERMINOLOGY AND EVENTS

Prior to presenting the many advantages of effecting control of anticoagulant therapy with the Prothrometer, it will be helpful to provide you with technical terminology applicable to the diseases requiring this treatment.

The main arteries supplying the heart tissue with blood form a sort of crown around the heart, and are therefore called coronary (crown) arteries, coronary vessels or just plain "coronaries." Blood does not normally clot within the arteries or vessels of the body. However, when a person has what the layman calls a "heart attack", a "thrombus" (clot) has formed and is plugging one of the coronary arteries. When a coronary artery is thus occluded (blocked), the area of heart muscle which derived its blood supply solely from this vessel dies. Thus, the occlusion (blocking) of this blood vessel causes the death (or infarction) of the heart muscle tissue.

The phrase coronary thrombosis describes the initial action - the forming of the clot; coronary occlusion describes the immediate result of the clot - the blocking of the artery; coronary infarction describes the eventual result of the blood supply blockage - death of some tissue; while the phrase myocardial infarction (myo=muscle, cardial=heart) nails down the specific tissue affected - the death of the heart muscle tissue. These just mentioned terms are all sometimes used loosely to describe the same general event.

Cerebral thrombosis is the condition of having a clot occur in an artery of the brain. The layman calls this a "stroke". This clot affects the brain the same as the clot in the coronary arteries affects the heart. The required blood supply is cut off from certain sections of brain tissue with disastrous effects (paralysis, etc.) on those parts of the body controlled by that area of the brain.

Another pair of terms requires clarification, thrombosis and embolism. Thrombus has already been defined as a clot and thrombosis is the condition of having a clot. An embolus, derived from a Latin word meaning "thrown in", is any body such as a blood clot which flows in from another part of the body into an artery and plugs in. This is often the process when an artery of the brain or of the lungs is blocked. The end result is the same, occlusion of the vessel and tissue damage from interrupted circulation.

Most coronary occlusions probably originate from clots which develop in the coronary vessels, i.e., they are the result of a thrombus. However, patients with coronary occlusions often "throw off" part of a clot into the circulation. When this clot lodges, it is known as an embolus. Emboli from the heart often lodge in the extremities, the lungs or the brain.

The total clinical picture of coronary occlusions and their sequence of events may include an original thrombus and secondary emboli. For this reason, the group of conditions which revolve around this clotting is known as thromboembolic diseases. Many research projects indicate that anticoagulation therapy serves

(a) to expedite recovery from a coronary occlusion,

(b) tends to prevent recurrences of occlusions, and

(c) decreases the incidence of embolic incidents elsewhere in the body.

Thus, anticoagulant therapy is effective in the whole gamut of thromboembolic disease.

There are one or two facts which relate significantly to the sequence of events following coronary occlusion:

(a) the heart must continue to beat and can't take time off to rest, and

(b) each of the coronary arteries supply rather circumscribed parts of the heart without overlapping a great deal.

It follows from these two facts that when a coronary vessel becomes occluded, the muscle cells in a part of the heart are left without nourishment and die, yet the whole heart keeps beating. The result of working an injured section of muscle is first - pain, next - diminished function. If the patient lives, the part of the muscle which gets no nourishment dies, and a scar is formed. It takes a few weeks for the scar to form, and so it is very important during the first weeks after a coronary occlusion that patients be kept as near absolute rest as possible.

What may happen in the acute attack is that there is a little bit of plugging which affects only a relatively small part of the heart muscle. However, clotting may continue backward in the artery to involve larger and larger areas of heart muscle.

III. THE CHEMISTRY OF BLOOD COAGULATION

The chemistry of blood coagulation is extremely complex. The fundamentals of the process can be outlined as follows:

When tissue is injured and bleeding occurs, an enzyme, thromboplastin, oozes out of the tissues and combines with blood. The critical agent in the blood which is acted upon by thromboplastin is prothrombin. Prothrombin is a protein which is formed in the liver, and for the synthesis of which Vitamin K is required. Vitamin K itself is formed in and absorbed through the gastrointestinal tract; it is then carried to the liver and in that organ plays an important role in the formation of prothrombin. When prothrombin, which is carried in the blood, meets thromboplastin from the tissues, a new substance, thrombin, is formed. (The formation of thrombin occurs only in the presence of ionized calcium, which is always a normal constituent of blood.)

There is another protein in the blood called fibrinogen. When thrombin combines with or acts upon fibrinogen, a substance called fibrin is formed. As the term suggests, fibrin forms in long threads which entrap other elements of the blood to form a clot.

The process which has been outlined is the one which occurs naturally when the body is injured and bleeding occurs. (Also, blood will clot when it is drawn from the vein and left in a bottle; however, formation of a clot under these circumstances takes a considerable period of time.) It is not natural or normal for blood to coagulate within the vessels (intravascular) of the body. When blood does coagulate intravascularly, something has seriously upset the chemistry of coagulation which will result in the undesirable situations described in Section II.

IV. ANTICOAGULANT THERAPY

Anticoagulant drugs increase the time required for the blood to clot and, therefore, reduce the chance for a recurrence of the undesired intravascular clot.

Heparin is the anticoagulant which is most commonly used in the initial treatment of the acute heart attack, since it goes to work immediately, and because its route of administration is intravenous or subcutaneous injections. Oral anticoagulants with a "loading dose" are begun at the same time as the heparin injection, but these drugs usually do not become fully effective for 36 to 48 hours. At that time, the heparin is discontinued and a daily maintenance dose of oral anticoagulant is given.

As the patient recovers from his acute attack of coronary occlusion, the question arises - what can be done to prevent another attack? It is self-evident that there is some characteristic of his arteries or the blood within them which is conducive to occlusion and infarction. The fact that there were conditions which were conducive to occlusion makes it seem likely that these conditions might occur again. And indeed, the statistics bear this out. The chances are very good that a second and third occlusion will occur and eventually one of them will cause death.

There is a great mass of data which indicates that the predisposition for myocardial infarction to recur can be strikingly and favorably altered by decreasing the coagulability of the blood. The study of Manchester, "The Prevention of Myocardial Infarction", in the Archives of Internal Medicine for December, 1957, is an example of evidence to this effect. Manchester studied 561 patients who had had one or more myocardial infarctions, for periods of from one to ten years. He had a control group which received nothing but ascorbic acid, and an experimental group which received anticoagulants. There was another small untreated group. The anticoagulant group had one-third the incidence of subsequent infarction and one-eighth the mortality of the comparable control and untreated groups.

What is necessary, besides taking anticoagulants, to produce these dramatic results? The essential maneuver is to regulate the dosage of anticoagulants. If insufficient anticoagulant is given, its effectiveness in preventing subsequent infarction is probably minimal. On the other hand, when too much anticoagulant is given, the patient may develop tendencies to bleed - from the stomach, from the gastrointestinal tract, into bruises, through the kidneys into the urine, and from minor injuries as well as between the brain and the skull following relatively minor head injuries.

It would be a simple matter if patients could be studied carefully in the hospital and have a dosage schedule of anticoagulant set up for them which they would then continue indefinitely. Unfortunately, this is impossible. Many factors, such as medicines and incidental illnesses may increase or decrease blood coagulability. The only way in which anticoagulant therapy can be properly maintained is to have repeated checks on coagulability of the blood. This means, in effect, that patients should have a test of prothrombin time at intervals of from several days to three weeks and often even daily, and whenever there is any significant change in their health status, or whenever any abnormal symptoms appear.

V. THE PROTHROMBIN TEST AND THE PROTHROMETER

Measurement of prothrombin time is a necessary adjunct to anticoagulation, either in acute hospitalized cases or in patients who are maintained on anticoagulant therapy for prophylaxis.

The time required for drawn blood to clot can be speeded up if a quantity of prepared thromboplastin is added to it, and this laboratory procedure is known as determining "prothrombin time" or accelerated clotting time. Whereas all tissues contain thromboplastin, the one ordinarily used in the test to determine prothrombin time is derived from rabbit brain which has been prepared by removal of its water and fat content by acetone. The choice of brain tissue is made because the brain, once it is stripped of its covering vessels, is relatively free of blood.

Older methods for determining prothrombin time use blood which is drawn from a vein. This "venous" blood is usually kept from clotting by immediately adding a substance such as oxalate, which removes calcium and thereby interrupts clotting. Then the blood is taken to the laboratory and centrifuged. The clear, straw-colored fluid which is separated from cells by centrifugation is called plasma. It contains prothrombin and fibrin, but no thromboplastin and no ionized calcium. The formation of fibrin from this plasma is made to occur by adding thromboplastin and ionized calcium. Although the prothrombin time of plasma can be measured with the Prothrometer, the usual method is to use a drop of whole blood from finger puncture. The use of whole blood rather than plasma was first described by Ziffren, et al, in 1940. In 1945, Manchester and Rabkin developed the whole blood method further, reporting it in Circulation for November of 1945 under the title of "The Control of Dicumerol Therapy in Myocardial Infarction by a Simple Blood Prothrombin Test."

Manchester's method made use of finger blood to which thromboplastin was added. No calcium needed to be added because whole fresh blood contains calcium (unlike plasma, from which it has been removed). However, the method has been questioned because the reaction between blood and thromboplastin was allowed to go on at room temperature, and the optimal rate of chemical reactions of this kind is related to temperature. In addition, the Manchester method depended on turning the slide back and forth and seeing strands of fibrin. The Prothrometer automatically controls the temperature of the reaction mixture, and the blood and thromboplastin is stirred so that the end-point is clearly indicated when fibrin is "fished" out by the clot hook.

The usual time elapsed between the addition of thromboplastin such as Simplastin(R) and the formation of fibrin in "normal" blood is between 11 and 14 seconds in all methods irrespective of whether plasma or whole blood is used. The principal factor which influences the figure found on "normal" blood is the thromboplastin. As this gets older, it gets weaker, and if a normal value in excess of that which the reagent manufacturer indicates is normal is found, the technique should be re-examined

and/or the thromboplastin discarded. It is important to remember that each reagent manufacturer indicates "normals" expected from their solutions, and although the usual "normal" obtained from the more popular kinds is as shown above, there are some brands with higher anticipated values. For example, one brand which currently has limited distribution in the United States, indicates expected "normals" of between 38 and 42 seconds.

When anticoagulants are administered, the rate of coagulation is decreased and the prothrombin time is increased. Many physicians who treat patients who have had coronary occlusions strive to maintain a prothrombin time of two to two and one-half times whatever normal value is obtained. Others feel that even one and one-half times normal affords generous protection. This is a point on which sales representatives should NOT make recommendations. Similarly, we would not recommend particular anticoagulants, although we should be familiar with those which are commonly used, such as Coumadin, Dicumarol, Tromexan, Sintrom, Heparin, and Prothromadin.

As is so often the case with advances in modern medicine, the use of anticoagulants involves certain special technical problems and potential risks. The dosage of an anticoagulant must be regulated so that the coagulability of the blood is decreased, but it must not be decreased to the point that hemorrhage occurs. Underdosage, in the light of present knowledge, is relatively ineffective. Overdosage runs the risk of hemorrhage.

As mentioned previously, control of anticoagulant dosage requires repeated measurement of the tendency of the blood to clot - the prothrombin time. Until the development of micro methods which may be used with the Prothrometer, it was necessary for patients on anticoagulant therapy to go to a laboratory every week or two and have a considerable amount of venous blood taken for analysis. Laboratory procedures are time-consuming and costly.

As a result of these difficulties, many doctors and many patients chose to forego the known advantages of anticoagulant therapy! With the Prothrometer, determination of prothrombin time has become a simple procedure. A tiny drop of finger blood provides the answer in a few minutes. Testing is done in the office, in the laboratory, at the bedside, or in the patient's home. Technical skill in making determinations can be acquired in an hour. Complicated laboratory procedures are eliminated.